

**STUDIES ON LEAF SPOT DISEASE OF DOLICHOS
BEAN (*Dolichos lablab*, L.) CAUSED BY
Alternaria alternata (Fries.), Keissler**



THESIS

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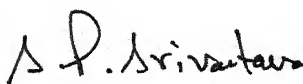
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CERTIFICATE

It gives me immense pleasure to certify that the thesis entitled "**Studies on Leaf spot disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler**" submitted by Sri Brajnath Pandey for the award of Degree of Doctor of Philosophy in Botany of Bundelkhand University, Jhansi is under my supervision and guidance. The thesis embodies the original research work of candidate himself and is fit for publication.

The present thesis, has been completed within the specified and prescribed time. He had put the required attendance in the Department during the period as per Ordinance of Bundelkhand University, Jhansi. I wish the candidate all success.


(Dr. S.P. Srivastava)

Dedicated to
My Beloved Sister
Late (Km.) Vimala Devi

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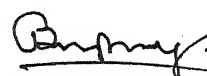
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Chapter - I
INTRODUCTION

INTRODUCTION

Plant diseases are the greatest hazards amongst the others in crop production. No agricultural commodity in world has exercised such an intensive influence on men. Vegetable crops have a prominent place in human diet as they not only adorn the table but also enrich the health of rural and urban population and save cereal and grains for maintaining the price line of food stuffs. They form the most himalyan health, resplendent redness, which is unfailing symbol of health, life cheerfulness and ecstasy. Vegetables are essential for a balanced diet and maintenance of good health. They supply carbohydrates, fats, proteins, vitamins and minerals, which are essential for the body.

India is predominantly being a vegetarian country, therefore vegetable yielding crop viz., *Abelmoschus esculentus* (Lady's finger), *Allium cepa* (Onion), *A. sativum* (Garlic), *Artocarpus heterophellus* (Jack fruit), *Capsicum annuum* (Chillies), *Carissa carandus* (Karonda), *Citrullus vulgaris* var. *fistulosus* (Tinda), *Cucurbita maxima* (Pumpkin), *Daucus carota* (Carrot), *Dolichos lablab* L. (Bean), *Lagenaria vulgaris* (Bottle gourd), *Pisum sativum* (Pea), *Solanum melongena* (Brinjal), *S. tuberosum* (Potato), *Spinacea oleracea* (Spinach), *Trigonella foenum-graceum* (Fenugreek) and *Brassica* sp. viz. *B. oleracea* var. *capitata* (Cabbage) *B. oleracea* var. *botrytis* (Cauliflower), *B. campestris* and var. *sarson* (Sarson), *B. rapa* (Turnip) etc. have a prominent place in our diet.

Dolichos bean (*Dolichos lablab*, L.) is one of the most important vegetable crop of the world. This crop is also known as Kidney bean, hyacinth bean, lab lab bean, Bonavist field bean, Hindi - Sem, Beng.-Shim, Guj. - Val; Mar - Pavta, Tel. - Chikudu, Tam. - Avarai, Kan. - Chapparadaavare and Mal. - Avaraa. Mostly it is used as a vegetable crop but sometimes also consumed as pulse, besides being a rich source of protein, minerals and vitamins for human beings. It also enriches in the soil due to fixation of atomospheric nitrogen by root nodule bacteria and has the unique quality of preventing soil erosion. A large population in India is vegetarian and a sizable part of which is suffering from protein malnutrition due to short supply and high price of pulses and protein rich vegetables, which have gone beyond the reach of poorman. Therefore every effort must be made to boost up the production of this important and common vegetable crop.

PLATE -I



Morphology of Dolichos bean (*Dolichos lablab*, L.)

Dolichos bean is a member of sub-family Papilionaceae of family Fabaceae (Leguminosae) and its cultivation can be traced back in the antiquity. Wild species of beans are found in India and probably India is place of origin (Choudhary, 1967). It is called as a sub-tribe Euphaseoleae under one of the main tribes viz., Phaseolae. It is a perennial, twinning or creeping herb and generally cultivated as annual leaves are pinnately trifoliate. Flowers are of various colours but generally are pink and white borne on axillary racemes. Pods are flat or inflated; linear or broad; 1-5 inch long with globose; ovate or flattened seeds varying in colour from white to dark black as given in Plate No. 1.

Plant is considered to be Asian in origin and distributed throughout the tropical and temperate regions of Asia, Africa and America. Two varieties are distinguished, one as annual commonly cultivated as a garden crop and other a perennial in varying degrees cultivated as a field crop. These varieties are indistinguishable in India and there has been a great difference in respect of characters and composition of the cultivated beans. The former is most common in Rajasthan, while later is most important pulse crop in parts of Madras and Mysore states.

Dolichos bean can be grown in both sub-tropical and tropical climate conditions. It is relatively a cool season crop but can be kept alive in summer to provide pods in the next season. However perennially, it gives poor pod yield and is advisable to grow only as annual. It is grown almost in all the States as dry crop. It can be grown in wide range of poor soils and is hardy and drought resistant. It may be grown on any type of soil except highly alkaline types. It thrives best on light sandy soil and is extensively grown on red loams, black cotton soils and stony and gravelly upland soils of Deccan. It is oftenly grown as preparatory crop on reclaimed lands. Optimum sowing is done between July and August. Seeds are sown in broadcast or in rows. It is grown as a Kharif crop mixed with Bajra and a rabi season crop in rice fields. The sowing time for pods is October - November in Bengal, for fodder is June, August or November or three times in the same field.

The seed rate is variable The rate is 40.0 lb per acre in South India, sown mixed with *Brassica nigra*, 20-25 lb. per acre in North India.

It is also grown as green manure crop particularly on lands newly borned under cultivation.

Seeds may be sown along the fringes of gardens and vines may be allowed

to trial on herbaceous plants. Copious and frequent irrigation is necessary. Flowering generally begins in November and pods are collected from December onwards up to March-April (Friminger, 1947).

Unlike many vegetables Dolichos bean, has variable practice of cultivation. It is grown as a field crop in Andhra Pradesh, Madras, Madhya Pradesh and Maharashtra. Contrary to this garden varieties are rarely found in Southern India, although common in Central, Eastern and Southern India. In North India particularly in U.P., it is mostly grown in kitchen gardens or thatches in the cities and villages.

India is the second largest producer of vegetables next to China. In India total area under cultivation of vegetable crops is 5.24 million hectares with production of 50,000 million tonnes (Anonymous, 2005). By the year 2000 A.D. Indian population is expected to one billion requiring more than 1000 million tonnes of vegetables. During 1991-1992 daily per capita consumption of vegetables in India was only 135.0 gms., which is much lesser than 285.0 gms. for a balanced diet (Majeed Gowda Nage, 1992).

In the world as a whole beans are french bean, cow pea, dolichos bean, sem, cluster bean and broad bean. They are cultivated almost in all the countries with an average production of 6.30 tonnes/ha., while the local varieties yield only 2-10 tonnes/ha (Anonymous, 1988).

Of the different types of beans, Dolichos bean is specially appreciated due to its flavour, colour and other characteristics. The pods in most types retain their tenderness, until they attain full size, when the seeds alone can be used. Flavoured types are those, which have a good flavour and thick fleshy skin with no fibre. Young pods may be salted or steamed and sun dried for preservation. Pods and seeds are also used as cattle feed (Burkill, 1935).

Dolichos bean surpasses most of the vegetables in its nutritive value. Fresh pods contains moisture 82.40 per cent; protein 4.50 per cent; fat 0.10 per cent; mineral matter 0.10 per cent; fibre 2.0 per cent; carbohydrates 10.0 per cent; Ca 0.05 per cent, phosphorus 0.60 per cent; iron 1.67 mg. and nicotinic acid 0.80 mg./100 mg. The vitamin content varies from 7.33 to 10.20 mg./100 mg. for uncooked samples and 0.77 to 1.12 mg./100 mg. for cooked samples as well as riboflavin 0.60 mg. (Anonymous, 1951 and Chaudhary, 1967).

Seeds contain moisture 11.80 per cent; protein 22.0 per cent; fat 0.50 per cent; mineral matter 3.10 per cent; fibre 5.30 per cent; carbohydrates 57.30 per

cent; calcium 0.28 per cent; phosphorus 0.39 per cent; iron 7.60 mg., nicotinic acid 1.50 mg. and carotene 119.0 per 100 gm. The globulins contain arginine 6.0-7.0 per cent; tyrosine 6.68 per cent and lysine 6.0 per cent, but are deficient in cystine and tryptophane. Germinated seeds and seedlings are a source of *l*-asparagine. They are also a rich source of urease.

The seeds are considered as febrifuge, antispasmodic and aphrodisiac (Nadkarni, 1927 and Kirtikar and Basu, 1935). The seeds are astrigent, diuretic and tonic (Chopra 1935).

The plant is a rich source of high nutritive and palatable fodder to cattle. The average composition of green feed and hay on dry matter basis is as follows. The green feed contains fibre 28.08 per cent; ether extract, 3.50 per cent; total ash, 14.80 per cent; CaO, 2.77 per cent; P₂O₅, 0.60 per cent; MgO, 0.97 per cent; Na₂O, 0.55 per cent and K₂O, 3.52 per cent. Hay on dry matter basis is made up of fibre 36.12 per cent; ether extract, 2.25 per cent; total ash 12.50 per cent; CaO, 3.78 per cent; P₂O₅, 0.60 per cent; MgO, 0.30 per cent; Na₂O, 0.75 per cent and K₂O, 2.40 per cent.

It is extensively used in South India as feed for cattle and horses in North India. The seeds are cooked before feeding. Stems, leaves and split husks are also used as cattle feed. It is used mainly as green pods but in dry form. The immature and mature pods are eaten and cooked as vegetable.

Seeds are consumed by poor classes after cooking or frying. They are eaten whole or after grinding into a meal. They are consumed as pulses after splitting. The foliage of the crop is also used for green manure, silage and hay in forage particularly for horses.

The facts narrated above reveal that *Dolichos lablab*, L. can play a dramatic role in the Indian agriculture economy by boosting the production and much of foreign exchange can be saved by eradication of diseases.

Among the various factors for such a poor yield plant diseases of varied origins taking away or heavy toll of the crop every year caused by pathogenic organisms (leaf spots), affected the germination, plant health, productivity and quality, which carry the inoculum rendering it unfit for consumption and affecting national economy. The foliar diseases are known to cause up to 20.0 per cent reduction in yield (Dastur and Asana, 1960). The production of *Dolichos* bean has become so much pampered, which presently requires to be set by increasing array of ills and pests by both skilled and persistent attention of the

agents causing diseases. Out of which fungi, bacteria, nematodes, mycoplasma and viruses are important. More lethal diseases and other pathogenic and no-pathogenic fungi associated with the seeds may cause serious damages.

The eradication of diseases, shall have to be given a prima importance through use of chemicals and genetic manipulations. If the ever burgeoning population of this country is to be sterred out of malnutrition, suitable protection and production technology must be evolved to agument the production of this important vegetable crop.

The cultural practices for disease control can also be adopted. Evolution of cheap and efficient pest and disease control measures based on integrated approach including genetic, chemical and biological methods can also be employed.

A keen intention of Plant pathologists, has been given on the fungal bacterial and viral diseases of this crop but inadequate information is available about the leaf spot disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.) Keissler. During the survey made in the year 2001 and 2002, the different varieties of *Dolichos lablab*, L., were recorded adducing symptoms of leaf spot caused by *Alternaria alternata*.

In view of seriousness of disease and being destructive in reducing productivity and importance of the crop as well as complete lack of detailed knowledge, it was thought necessary to take up the detailed study of the "Leaf spot disease of Dolichos bean (*Dolichos lablab*, L.), caused by *Alternaria alternata* (Fries), Keissler, it is imperative to work out the detailed study of disease and its pathogen as well as to suggest control measures to get rid off the disease, the present investigation was therefore undertaken with the following objectives -

1. Survey and collection of diseased material from the different locations of U.P. and to study the prevalence, severity and symptoms of disease.
2. Isolation, purification and pathogenicity test of pathogen.
3. Identification of the pathogen on the basis of morphological and cultural characters.
4. Production of cell wall degradation enzymes in *vivo* and *vitro* by the pathogen.

5. Studies on the nutritional requirement of the pathogen and effect of temperature, pH and N.P.K. on it.
6. Susceptible growth period of host and study of climatic conditions influencing the development of the disease.
7. To find out the effect of pathogen on certain biochemical constituents of diseased parts of host.
8. Investigations on mode of perpetuation and spread of diseases.
9. Screening of Dolichos bean germplasm on disease resistance.
10. Studies on host range of the pathogen.
11. Bio-asssay of different fungicides against the pathogen in *vitro* and efficiency of the effective ones in controlling the disease in *vivo*.

□□□

Chapter - II

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Bean (*Dolichos lablab*, L.) being an important vegetable crop of India and foreign countries, has attracted the attention of Plant Pathologists all over the world. It suffers from a number of diseases caused by Fungi, Bacteria, Viruses and Nematodes, which cause deterioration of quality and reduction in quantity of the production to a great extent. Amongst fungal diseases are Alternaria leaf spot, Anthracnose, Powdery mildew etc., commonly found on the plant and various other pathogenic and non-pathogenic fungi associated with the seeds excel others in numbers and are destructive.

Alternaria leaf spot of Bean caused by *Alternaria alternata* is comparatively a new disease observed for the first time in Rajasthan in 1966. Later Singh *et al.* (1992) from Jammu and Kashmir reported a new leaf spot disease of Field bean caused by *Alternaria alternata*. Barrus (1911, 1915, 1918 and 1921), Leach (1923), Reddick (1923), Andrus and Moore (1935), Yu (1937), Andrus and Wade (1942), Ramanowski *et al.* (1962), Goth and Zaumeyer (1965), Master Brock (1980) and Menezes and Dianese (1988), studied the prevalence of Bean anthracnose caused by *Colletotrichum lindemuthianum*. Bean powdery mildew was studied by Pauvrt (1991). Vallejos (1988), reported *Macrophomina phaseolina* causing Root and Stem rot. A few internal seed borne fungi viz. *Fusarium semitectum* and *Phomopsis* sp. were also isolated by Dhingra (1978).

A little work has been done on the leaf spot of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler except the report of occurrence. No information is available on etiology, mode of perpetuation, sources of resistance, chemical control of disease etc. The work done of some other crops in relation to Alternaria leaf spot caused by *Alternaria alternata* sp. is being reviewed here to supplement the information.

TAXONOMY AND NOMENCLATURE OF THE PATHOGEN :-

Alternaria alternata, is represented by a number of isolates ranging from saprophytes to parasites having polyphagous nature. Due to this wide prevalence in nature several workers have studied the morphological and cultural characters of the fungus, *A. alternata*, since the identification of single species of *Alternaria tenuis* by Nees in the year 1817, during the course of his studies on the genus *Alternaria*. Thereafter a number of species have been added to Genus *Alternaria* and at present is comprised of one hundred species

parasitizing numerous cultivated plants and weed plants producing necrotic leaf spots.

Fries (1832), in his "Systema mycologia", described *Torula alternata* and kept Nees species, *Alternaria tenuis* as its synkaryon. The correct name of the pathogen became *Alternaria alternata* (Fries.), Keissler, which is generally accepted today by most of the Plant Pathologists.

Elliot (1917), after a century of first discription of *Alternaria* by Nees (1817) identified the form of conidia as obclavate, pointed often with a long beak as a genetic criterion and recorded that chain formation might be suppressed during unfavourable conditions. Bolle (1924) and Nola (1927), also recognised Elliot's concept of the genus by isolating a fungus from brassicaceous and other plants and identified as *A. tenuis* with very short beaks.

The nomenclature of *A. tenuis* since long been the matter of discussion amongst the Mycologists (Mason, 1928, Wiltshire, 1933 and 1938; Groves and Skolko, 1944; Neergaard, 1945; and Simmons, 1967 and Lucas, 1971).

Wiltshire (1933 and 1938) made the studies on the concept of *Alternaria* and *Macrosporium*, who was of the opinion that *Macrosporium* should be suppressed in favour of *Alternaria*, typified by *Alternaria tenuis*, Nees, the type species of which Wiltshire was unable to locate the examination.

Groves and Skolko (1944), described the taxonomy and morphology of some of the species of seed borne *Alternaria* sp.

A monographic study of the genus, *Alternaria* with reference to the taxonomy, parasitism and economic significance was worked out by Neergaard (1945) and the systematic study of the different groups of fungi made by him is one of the best work so far produced.

Jolly (1959), discussed the morphological variations of species in the genus, *Alternaria* and thereafter he also made the monographic study of the genus, *Alternaria* in 1964 and framed a very simple key for identification of the most common species dividing them into three sections.

Alternaria tenuis, Nees described by Nees in 1817, as a type species of the genus but its description was not sufficient to draw a positive identification of the species itself. *Alternaria tenuis* is an invalid name due to prestarting date epithet (Simmons, 1967).

Simmons (1967), was successful in obtaining an authenticated specimen of the type species of *Alternaria*, Nees in Persoon's herbarium and proposed it as a neotype of *A. tenuis*, Nees, which was accepted by Ellis (1971) and Lucas (1971) and the name of *Alternaria alternata*, has come in wide use after the publication of typification in the genus, *Alternaria* by Simmons (1967).

Lucas (1971), suggested the name of *A. longipes*, is not needed and should be placed in synonymy with *A. tenuis* for which according to internal rules of Botanical Nomenclature the valid name is *A. alternata*.

The genus, *Alternaria* is placed in the family-Dematiaceae belonging to class Deuteromycetes. Characters and classification of *Alternaria* sp. has also been described by Subramanian (1971), in the monograph of Hyphomycetes.

The suggestions given above with reference to taxonomy were later accepted by the Mycologists like Tweedy and Powel (1963); Siddiqui (1963) and Rao (1971).

Several species of *Alternaria* viz; *A. alternata*, *A. gossypina*, *A. macrospora* and *A. tenuis* were also reported by Faultwetter (1918), Young (1929), Hopkins (1931), Rao (1966), Simmons (1967), Yousef *et al.* (1971), Kamal *et al.* (1971), Chauhan *et al.* (1972), Chauhan and Suryanarayana (1972), Srivastava (1976), Chopra and Sharma (1976), Vinayagamorthy *et al.* (1976), Padmanabhan and Narayanaswamy (1977), Desai and Patil (1982), Padaganur and Basavaraj (1988), Bashi *et al.* (1983), Singh *et al.* (1984), Khedi *et al.* (1986), Cooty (1987) and Krishna Mohan and Vidyasekaran (1989).

SYMPTOLOGY :-

Bose (1942), first time recorded the pathogenic attack of *Alternaria alternata* (*A. tenuis*, Nees) (Fries.), Keissler on the leaves of *Helianthus annuus*, L. causing leaf spot disease from India. The symptoms first appeared on the upper surface of leaves as small and oval spots. Later, numerous such spots coalesced together and covered maximum surface of lamina. Sometimes affected areas of leaves were perforated irregularly due to falling away of dead tissue, resulting into "shot holes". In general the mature plants showed the symptoms of the disease on the upper surface of leaves.

The leaf spot and blight disease of Pea caused by *Alternaria alternata*, was studied by Agrawal (1961). The disease appeared in the form of pale to light brown spots on leaves. The spots increased in size, became circular to irregular,

dark brown, which later on coalesce and infected portion dried up and withered away.

Goyal (1968), observed the *Alternaria* leaf spot disease of *Dolichos* bean caused by *Alternaria alternata* as a new and serious disease from Rajasthan, India. The disease appeared in the form of small, brown, circular to irregular pustules on leaves arising from the margin. In severe attacks the pustules were coalesced, which lead to blighting of lamina and finally defoliation. He also established pathogenicity of *A. alternata* of *Dolichos* bean.

The infection of *A. alternata* on Cluster bean was observed by Chand and Verma (1968) in India.

Aulakh (1969), observed *Alternaria* leaf spot disease of *Arachis hypogea* caused by *A. alternata* in Punjab. The disease symptoms appeared as brown, water-soaked, translucent and blighted areas on tips of leaflets, which increased in size until 30-40% of leaf was affected.

Gupta (1970), reported a new leaf spot disease of Mungbean due to *A. alternata* from Udaipur and reported the incidence of the disease to a tune of 80.0 per cent with leaf area infection of about 45.0 per cent.

Singh (1971), described the symptoms caused by *A. alternata* on the leaves of *Saccharum officinarum* as elliptical to oval or cylindrical lesions parallel to long axis but were found as dark brown with reddish brown bands towards the margins. The area between lesions became chlorotic and sometimes the entire leaves were found dried.

Gonzalez (1973), studied the leaf spot of Bean (*Phaseolus vulgaris*) caused by *Alternaria* sp. and characterized by irregular round and light brown lesions with darker margins, which coalesce and cause leaf shedding.

Graud *et al.* (1977), observed the *Alternaria* leaf spot on Hyacinth bean (*Lablab purpureus*) caused by *Alternaria* sp., The primary symptoms appeared on the leaves as small flecks of reddish brown colour. As the disease advanced, the flecks turned into round spots with black border. Later the affected portions fall off resulting in "Shot hole" symptoms. Sometimes the scorching effect on the leaf margin of the affected leaves was also recorded.

Gurha *et al.* (1981), recorded first time the prevalence of pathogenic attack of *Alternaria alternata* on Broad bean (*Vicia faba*, L.) and searched the sources of resistance against the pathogen.

Sharma *et al.* (1983), observed new Fruit rot on Kidneybean (*Lablab purpureus*, L.) caused by *Alternaria tenuissima* in which fungus caused 20.0 per cent rot of fruit tissues within 10 days. The rottened tissues turned black, soft, formed a shallow cavity and emitted pungent odour with mycelium covering the infected area.

Singh *et al.* (1984), reported *A. alternata*, parasitizing the leaves of *Gossypium arboreum* plants and was found as a new record from India causing leaf spot of Cotton.

Gupta (1985), recorded the occurrence of leaf blight disease of Broadbean (*Vicia faba* L.) caused by *A. alternata*. The disease started from the tip or the margins of the lower leaves, which gradually increased and covered the entire leaf area. The disease first appeared in the month of November on the young plants. In severe cases the entire foliage was blighted and defoliated.

Later Gupta and Basuchoudhry (1986), reported leaf blight of Broad bean caused by *A. alternata* in Manipur.

The pathogen, *A. alternata* is widely distributed and prevalent on the various crops by producing different symptoms on various crops viz; Linseed (Siddiqui, 1963); Rape (Vaartnou and Tewari, 1972 and Narain and Saksena, 1974); Arhar (Mehta and Sinha 1982) and Cotton (Rao, 1965).

It is quite obvious that literature discussed above that no detailed work appears to has been done on the leaf spot disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, except the reports collected from different parts of world and detailed information is available on *A. alternata* in relation to physiology, enzyme production, host range, survival, spread effect of climatic conditions and effect of fungicides to arrest its growth.

PATHOGENICITY TEST :

Bose (1942), carried out inoculation experiment with *Alternaria alternata* using both injured and uninjured leaves of *Helianthus annuus*, L. and found leaf spot symptoms developed within four to ten days respectively.

Kamal (1950), established the pathogenicity of *A. tenuis* on *Pandanus* sp. and after inoculation proved its pathogenic nature although a week one and infection induced only on wounded leaves.

Tandon and Chaturvedi (1965), studied *Alternaria* leaf spot of *Lycopersion*

esculentum, caused by *Alternaria alternata* and observed in pathogenicity test that it was a weak parasite of Tomato and can penetrate only wounded leaf surface.

Vander Ende and Van Hock (1966), found that after inoculation with a spore suspension of *A. alternata* the inner epidermal cells of the endocarp variety undusa bean (*Phaseolus vulgaris*, L.) exhibited a specific pattern of ectodesmata changing as the infection hyphae developed.

Stavely and Slana (1971), observed that *A. alternata* penetrated inoculated older leaves of *Nicotiana tabacum*, directly causing necrosis of epidermal cells and invading the mesophyll cells intercellularly. Mechanical wounds of young leaves elicited a similar response.

Drobey *et al.* (1984), recorded that *A. alternata* infected the Potato leaf by direct penetration and through stomatal openings. A differential susceptibility of leaves, was observed in which the middle leaves of plant showed the highest disease incidence at given growth stage.

MORPHOLOGICAL AND CULTURAL CHARACTERS OF THE PATHOGEN

(a) MORPHOLOGICAL CHARACTERS -

Alternaria, is a well known for its variability and polymorphic nature (Mason, 1928; Groves and Skolko, 1944; Neergaard, 1945; Simmons, 1967 and Lucas, 1971 and 1976). The fungus is characterised by dark coloured, obclavate and muriform conidia but somewhat variable in respect of size, septation and length of beak borned in long chains.

Mason (1928), recorded that majority of spores of *A. alternata* produced in culture had 3-5, 11 transverse septa and measuring 20-50x 10-14 μ a statement with which Groves and Skolko (1944), concur and they quote the spore measurement as (16)-20-50(70) \times (7)-10-16(20) μ and observed in nature the beak may be proportionately longer.

In 1933, Dey studied the morphology of fungus, *A. alternata* causing Blight disease of *Linum usitatissimum*, L. and observed that mycelium was pale olive to dark in colour and second cell of spores from below had the largest diameter constricted, 3-7 celled, measuring 10-40 x 5-10 μ m. with beak (average 24x7 μ m.) and beak 3-7 μ m. (average 4.9 μ m.) No difference in size of conidia was observed.

Bose (1942), described the character of *Alternaria tenuis* isolated from infected leaves of *Helianthus annuus*. The mycelium was septate and hyaline, when young, changing into brown later. The conidiophores were simple; sometimes branched; septate; straight or bend and light brown in colour. In culture the conidia were usually singly borne, sometimes in chains of 2-4 but catenulation was more common in old cultures. They were ovate, sometimes clavate with a slender base and rounded apex. The conidia were variable in form as well as in size but generally were broad and muriform. The number of transverse walls varied from 3-4 but generally from 4-7.

Neergaard (1945), made the study of several isolates and found that the spores have up to 9 transverse septa and total length of spore including beak varied from 7.0 μ m - 72.0 μ m. He studied the morphological and cultural characters of 40 isolates of *A. alternata* and recorded a wide variation in growth and size of conidia establishing the fact of being polymorphic in nature.

The fungus, *A. alternata* produced cottony mycelium, which gradually developed a colony with peculiar alternate zonation of white and grey colour on attaining full size. The colony became deep black due to preponderance of conidia and was velvety in appearance. The vegetative hyphae were hyaline, septate and filamentous. Conidiophores arise in culture medium on hyphal branches and were olive brown in colour. The conidia were found in chains of 7-9; olive brown in colour; beaked; obclavate and long to oval in shape, measuring 8.0 - 78.0 \times 6.0-22.0 μ . Both transverse and longitudinal septa were present and were usually 5-7 celled. Beaks were short and false having the same colour as of spore body (Kamal, 1950), while Arya and Prasad (1952), observed that conidia formed in culture medium were smaller (10.20 - 40.50 \times 5.0 - 13.50 μ) than those produced on host (16.70 - 45.50 \times 7.0 - 16.70 μ).

Malone and Musket (1964), made a detailed study of 10 cultures from seed coat developing on 2.0 per cent Malt extract agar medium and observed that conidiophores were brown; simple or branched; variable in length; septate and 3.0-6.0 μ wide. The conidia were formed in long; simple or branched chains; brown; generally smooth walled; sometimes rough-walled; obovoid or obclavate, muriform with average 4-5 transverse septa and several longitudinal septa with constriction at the septum measuring 16-(37.2)-80.0 \times 8-(13.1)-24.0 μ . Branching of chains generally took place at the beak of spore with several scars and became geniculated. Sometimes the branch chains develop from a short lateral projection of the beak. The colonies on the medium showed a pronounced

wide range in regard to quantity of aerial mycelium, colour of colonies and sporulation. The aerial mycelium at one extreme end might be scant, whereas at other end it was observed as dark and floccose. The colonies also vary in colour, generally being a shade of grey but may also be white and olivaceous brown or almost black. Usually spores were found either few in numbers or numerous.

(b) CULTURAL CHARACTERS -

Besides morphological characters many Plant pathologists have also studied the cultural characters of the pathogen. Lilly and Barnett (1951), suggested the superiority of natural media over synthetic media in relation to mycelial growth of fungus due to the fact that former contains more nutrients than the later. No distinction was recorded regarding sporulation. Hawker (1956), also recorded that mycelial growth was favoured by nutritional factors but was not found necessary for maximum sporulation.

Swank (1951), recorded that on Potato dextrose agar (P.D.A.) medium the aerial mycelium of *A. alternata*, was cottony and dark grey. The conidia were found $28.70 \times 12.30 - 20.50 \mu$ with 4-7 transverse septa and 0-3 longitudinal septa. The beak was $20.50-73.80 \mu$. long with 0-4 transverse septa and total length of spore was $49.20-15.40 \times 12.30-20.50 \mu$.

Arya and Prasad (1952), found maximum growth and sporulation of *Alternaria brassicae* and *A. macrospora* (*A. alternata*) on linseed on host leaf extract.

The pathogens, *A. alternata* and *A. helianthi* were observed as fast growing on Potato dextrose agar medium followed by Richard's medium at the optimum temperature of 25°C . (Ashour and EL-Kadi, 1958 and Allen *et al.* 1983).

Kapoor and Hingorani (1958), discussed the morphological characters of *A. alternata* isolated from leaf spot and Fruit rot of *Solanum melongena* on Potato dextrose agar (P.D.A.) medium showing zonations of dark olive colour alternating with pale olive areas except near the periphery, where it remained whitish. The hyphae were observed as dark olive buff to buffy brown; septate; $3.60-7.0 \mu$ wide; conidiophores septate, $40.0-70.0 \mu$ and $3.0-6.0 \mu$ wide; erect; simple or branched; geniculated; conidia formed in chains, usually made up of 7-10 spores; smooth when young; linear to obclavate, dark olive buff in colour; 1-7 transverse septa and 0-5 longitudinal septa and measuring $20.0-28.0 \times 7.0-18.0 \mu$. The beak of the conidium was made of apical cell of the spore body alone.

Rangaswami and Sambandan (1960), recorded that Potato agar medium supported maximum growth of six isolates of *Alternaria* sp.

In synthetic media, Richard's medium was found to be better for growth and sporulation of *Alternaria* sp. (Rangaswami and Sambandan, 1960 and Goyal, 1977). Rangaswamy and Chandrasekaran (1962) and Chopra and Jhooty (1971), also found that semi-synthetic media supported better growth than synthetic media.

Effect of culture medium on the morphological characteristics of the pathogen have also been studied by Neergaard (1945) and Gupta *et al.* (1969), who observed that cultural characters of spore morphology of *Alternaria* sp. were influenced by the ingredients of substrate media.

Siddiqui (1963), in his studies with fungus causing Blight disease of many plants, found no difference in size of spores in culture and those on host and observed that *Alternaria lini* Dey, *A. burnsii*, Uppal, Patel and Kumar; *O.A. citri*, Pierce; *A. palsdui*, Ayyanger; *A. chartarum*, Preuss and *A. humicola*, Oudeum, which were described under different names should be grouped with *A. alternata*.

Gupta and Uprety (1964), reported better growth and sporulation of *Alternaria tenuis* on decoctions containing Tamarind (*Tamarindus indica*), *Cassia fistula*, *C. obtusifolia* and *Mangifera indica* than of Potato decoction. Wide variations in morphological and cultural characters of *A. alternata* isolated from different hosts were also studied by Dhanraj (1970); Reddy and Bilgrami 1974; Ram and Gupta, 1975; Madaun *et al.* 1979 and Mekani, 1980.

Sokhi *et al.* (1972), studied the cultural and morphological characters of *A. alternata* isolated from wheat (*Triticum vulgare*) leaves and recorded the presence of bigger spores on hosts as compared to those obtained on Standard nutrient and Malt extract agar media.

Lima bean agar medium was found as best medium for the pathogen, *Alternaria alternata* by Ionnaidis and Main (1973), while Reddy and Bilgrami (1974) observed variation in morphological and cultural characters of different isolates of *A. alternata*.

Mathur and Sarbhoy (1977), made cultural studies of *Alternaria alternata* infecting Sugarbeet with particular reference to selection of a basal medium. Maximum growth of the isolate was recorded on Richard's medium, followed by

Czapek's (Dox), modified Czapek's (Dox), modified Brown's and Asthana and Hawker's media.

Graud *et al.* (1971), observed that Coons liquid medium, Potato dextrose and Host extract with 2.0 per cent dextrose proved the best media for conidial germination of *A. circinans* (*A. brassicicola*) and Hyacinth bean (*Lablab purpureus*, L.).

Chandrashekhar and Ball (1980), observed that *A. alternata* grew satisfactorily with its maximum growth on 2.0 per cent Malt extract agar medium, but Xu *et al.* (1984), observed rapid mycelial growth on Potato dextrose agar medium, which was supported by Susuri and Hagedorn (1986), who found the same results.

Mohanty *et al.* (1981), reported that the growth and sporulation of the fungus, *A. carthami*, were best on Richard's medium.

Reddy and Gupta (1981), tested six liquid media in which Potato dextrose was the best for growth and sporulation of *A. helianthi*.

Chattannavar *et al.* (1987) isolated, *Alternaria alternata* from infected leaves of *Triticum vulgare*, L. and purified it by single spore culture technique and reported that isolates differed in their growth on different media and maximum growth was observed on 12th day on Potato dextrose agar medium.

Patil *et al.* (1988), observed that growth of *A. tenuissima*, was best on Sabouraud's agar among the solid media and on Richard's medium among the liquid ones tested.

Andre-Rodrigues and Lupuli-Santana (1993), found that high growth rates of *A. alternata* (a common pathogen of Potato and Tomato), were achieved on the new media yucca glucose agar and Malanga glucose agar tested.

PATHOGENICITY TEST

Many plant pathologists have established the pathogenicity of *Alternaria alternata* on different species of *Helianthus annuus*, L. Bose (1942), isolated *A. tenuis* in both injured and un-injured leaves of Sunflower and recorded the development of symptoms of leaf spots within an incubation period of 4-10 days.

Kamal (1950), also got success in establishing the pathogenicity of *A. tenuis* on *Pandanus* sp. and after inoculation proved its pathogenic nature although a weak one and infection induced only on wounded leaves.

Tandon and Chaturvedi (1965), worked out *Alternaria* leaf spot of Tomato caused by *A. tenuis* and proved its pathogenicity but a weak parasite was able to penetrate only wounded leaf surface.

The older leaves of Tobacco inoculated with *Alternaria*, were found deeply penetrated causing necrosis of epidermal cells invading mesophyll tissues intercellularly. Mechanical injuries on young leaves elicited a simpler response (Staveland and Slana, 1971).

PHYSIOLOGICAL STUDIES (EFFECT OF TEMPERATURE AND HYDROGEN-ION CONCENTRATION) -

Temperature plays a significant role and sporulation of fungi. In the same way hydrogen-ion concentration (pH), determines the acidity or alkalinity of media especially a relevant factor for the development of fungal growth. It determines availability or non-availability of the nutritional requirements and elements from the media necessary for the development and growth of fungus.

(a) EFFECT OF TEMPERATURE ON GROWTH AND SPORULATION OF THE PATHOGEN -

The role of temperature on growth and sporulation of *A. alternata*, has been studied by several workers.

Neergaard (1945), observed that the optimum temperature for the growth of Danish isolate of *A. alternata* was 26°C. However Kamal (1950), from India reported 25°C as optimum temperature for its growth. Kapoor and Hingorani (1958), recorded the minimum, optimum and maximum temperatures for the growth of *A. alternata* were, 4°C, 28°C-29°C and 45°C respectively. Subsequently Ashour and El-Kadi (1958) recorded best growth of fungus at 25°C on Richard's medium.

Arya and Prasad (1962), observed the effect of temperature range of 10°C-35°C with optimum growth at 25°C, necessary for the growth of *Alternaria brassicicola* var. *macrospora*.

Better growth and sporulation of *A. tenuis*, was observed by Tandon (1961) at pH 5.0. Siddiqui (1963), recorded the temperature range of 12°C-30°C with optimum at 26°C. *A. tenuis* could grow with no growth below 7°C and above 35°C.

Verma (1963), observed the optimum temperature of 25°C was essential for

the growth and sporulation of *A. tenuis* (*A. alternata*). The optimum for the fungus was of 20°C–25°C in culture and laboratory (Crissan and Mesecu, 1970).

Csyzewska (1970), obtained fungal growth of *Alternaria tenuis* (*A. alternata*) (Structures A, B and C) at 0.5°C – 40°C (optimum 25°C and 27°C). Abundant sporulation occurred at 17°C – 35°C; 20°C – 30°C and 12°C – 30°C respectively. Kamal *et al.* (1971) and Girgarju (1976) studied the spore germination of *A. tenuis* (*A. alternata*) at 27°C (optimum temperature). The maximum fungal growth and sporulation of two isolates of *A. alternata*, was recorded at 30°C (Mathur and Sarbhoy, 1977).

Xu *et al.* (1976) observed that temperature of 26°C–28°C is required for germination of conidia of *A. tenuis* (*A. alternata*). Two isolates of it were observed as growing well on Potato dextrose agar (P.D.A.) medium at 20°C–30°C, (optimum 28°C). Maximum growth of fungus was obtained at temperature of 20°C.

Sheir *et al.* (1982), in their studies found that optimum temperature for growth of *A. alternata* was 30°C.

Susuri and Hagedorn (1986), reported that *A. alternata* grew well at the temperature range of 20°C–32°C with optimum at 28°C.

Chhattannawar *et al.* (1987), obtained maximum growth of *A. alternata* at 20°C.

Vakalounakis and Malathrakakis (1988), found that mycelial growth of *A. alternata* occurred at 5°C–40°C with optimum temperature of 25°C and spore germination occurred at 10°C–37°C with optimum at 26°C.

Shanda (1995), found that optimum temperature for the growth of *A. alternata* on melons ranged from 25°C–30°C.

(b) EFFECT OF HYDROGEN ION CONCENTRATION ON GROWTH AND SPORULATION OF THE PATHOGEN -

Comparatively few workers have investigated the role of pH on the growth and sporulation of *A. alternata*.

Ashour and El-Kadi (1959), observed that best growth and sporulation of *A. alternata* at pH 6.0.

Arya and Prasad (1962), observed that *Alternaria brassicicola* var. *macrospora*, can grow on a wide range of pH ranging from 3.0–8.50 with

optimum growth at the range of 5.60-6.50.

Verma (1963), observed that optimum pH 6.60 was essential for the growth and sporulation of *A. tenuis* (*A. alternata*).

Saad and Hagedorn (1970), was of the view that minimum, optimum and maximum pH for the growth and sporulation of *A. alternata*, were 4.40, 6.50 and 7.60 respectively.

The maximum fungal growth and sporulation of two isolates of *A. alternata*, was recorded at pH 5.50 (Mathur and Sarbhoy, 1977).

Xu *et al.* (1976), recorded that pH 5.60 is required for germination of conidia of *A. tenuis* (*A. alternata*). Two isolates of it were observed at pH range of 3.70-7.10 (optimum 6.50). Maximum growth of fungus was obtained at pH 6.50.

Singh *et al.* (1980), found that maximum mycelial growth and sporulation of *A. alternata* occurred at pH 5.50.

Mohanti *et al.* (1981), reported that both the growth and sporulation of the fungus, *A. carthami*, were best at pH 6.0.

Xu *et al.* (1984), while studying pH requirement of two isolates, *A. alternata* noted its growth at pH range of 3.70-7.10 with optimum of 6.50.

Chhattannawar *et al.* (1987), obtained maximum growth of *A. alternata* at pH 6.50.

PRODUCTION OF CELL WALL DEGRADATION ENZYMES IN VIVO AND VITRO -

Fungi, Bateria and Nematodes cause disintegration of tissues through the action of enzymes secreted by them. These enzymes are of different types for producing different actions affecting different tissues and chemical constituents of cell walls. Brown (1965); Bateman and Miller (1966) and Bateman and Basham (1976), described the role of enzymes produced by fungal species and their mode of action on host cells "Macerating principle" and "Lethal principle" was adopted by Brown (1965) for the purpose of discussion. According to him under the new system of classification the Macerating principle (Protopectinase) may be equated with either or both of the chain splitting polygalacturonase (PG) and polymethyl galacturonase (PMG) enzymes acting on middle lamella and more or less methylated macromolecular chains of pectin material in cell wall proper respectively.

The structure of framework of a plant cell is mainly composed of pectic and cellulolytic substances (Padmanaban and Narayanaswamy, 1978). The cell wall degradation is primarily carried out through the extracellular enzyme secretion by pathogens. (Sadasiwan and Subramanian, 1963; Wood, 1966; Bateman and Miller, 1966 and Agrawal and Hasija (1978), studied the involvement of pectic enzymes in degradation of pectic constituents of cell-wall and of the middle lamella in plant tissues including diseases eg. soft rots, dry rots, wilts, blights, leaf spots etc. The pathogenesis was caused due to the actions of pectic enzymes as one of the most complex factors was detailed as in numerous early findings made by Sadasivan and Subramanian, 1963; Brown, 1965; Mahadewan, 1967; Wood 1967, Alburshin *et al.* (1969), Bateman and Basham (1976); Mussel and Strand, 1972 and Agrawal and Hasija, 1978).

In certain diseases the pectic enzymes appear to be a principle factor, whereas in others they are of little or of no importance. Pectic substances are Colloidal Carbohydrates composed of linear polymeric chains of D-galacturonic acid alongwith α -1-4 glycosidic linkages (Kertesz, 1951). The free Carboxyl group is esterified with methanol (pectinic acid and pectin) or non- esterified pectic acid (Bateman and Miller, 1966). Cellulose is the major constituent of plant cell mainly in cell-walls. It is a linear polymer of D-glucose units with 1, 4 linkages. Celluloses secreted by parasites are important in pathogenesis being hydrolysed due to which cell walls loose their strength and then collapse.

Alternaria alternata decomposes the fibres by affecting pectin and cellulose (Ruschman and Bartram, 1940). Marsh *et al.* (1947) observed the decomposition of cellulose by *Alternaria* sp. Coulson and Mars, (1952); Hamcock *et al.* (1964); Heath and Wood (1971); Dour and Gayat *et al.* (1978) and Wood (1985), observed the rapid decomposition of Cellulolytic and pectic substances of leaves affected with *Alternaria alternata*, due to secretion of cellulose and pectic enzymes by the fungus during the existence as a saprophyte. Fischer (1953), studied the laboratory tests and decomposition of lignin by *A. alternata*.

The production of cellulase, pectinase and proteolytic enzymes by *A. tenuis* was established by Tandon and Srivastava (1950). *A. alternata*, produced maximum amount of cellulose in the substratum supplemented with pectin and cellulose powder than in carboxymethyl cellulose filter paper and glucose alone (Pandey, 1965). Later studies were made on the production and activity of extracellular enzymes in synthetic media by different species of *Alternaria* by Pandey (1965); Verma (1971); Marimuthu *et al.* (1974) and Mehta *et al.* (1975).

Kanavaskaya (1968), observed the decomposition of methylcellulose by *Alternaria alternata*. Later Prasad (1967), studied the effect of thirteen compounds on production of enzymes by the fungus and obtained Nepthol, Oxidised pyrogallol, Tannic acid and extract of heart wood of *Acacia catechu*, which inhibited activity of pectinase, and polygalacturonase enzymes, P-benzogquinone, hydroquinone and 1, 2, 5, 8-tetra hydroxyquinone were also found in culture media as reducing enzymes produced by the fungus, *A. alternata* after a period of inactiveness prior to beginning of inhibitory action.

Egorov *et al.* (1971), studied the formation of proteolytic enzymes by *Alternaria alternata*, supplied with lactose and maize extract. Hasiza (1974) found pectin to be effective in inducing production of enzymes.

Mehta *et al.* (1974 and 1975), found best seeretion of cellulolytic and pectolytic enzymes produced by *A. solani* and *A. tenuis* in synthetic medium after two days of inoculation. Thy also observed that these fungi produce polygalacturonase and Pectin methylpolygalacturonase (PMG), which played an important role than trans-eliminases in pathogenesis by *A. solani*. A high cellulose activity was also observed in culture filtrates by these fungi. Later in 1975, the best production of enzymes was studied by *A. alternata* and *A. solani* on eleven culture media and was found synthetic medium as best after 12 days of inoculation. Both the fungi produced glycosidases more effectively than trans-eliminases. Marimuthu *et al.* (1974), reported that in *vitro*, *Alternaria sesami*, produced cellulase, protease and macerating enzymes.

Mehta and Mehta (1974), recorded the effect of different compounds on the production of polygalacturonase trans-eliminase (PMTE) by *A. solani* and *A. alternata* isolates in *vitro*.

In another study Mehta *et al.* (1975) studied the best production of Pectolytic enzymes produced by *A. solani* and *A. tenuis* in synthetic medium influenced by twelve days incubation period and observed that these fungi produced glycosidases more effectively than transeliminase under the same conditions. They also recorded that 12 days incubation period was found optimum for production of carbomethylcellulose on glucose source, while cellulose production was found best at pH 4.0. Both the fungi were also found to be termed as cellulolytic since they mecerated both C1 and Cx type cellulolytic enzymes (Reese, 1963). Cellulolytic enzymes act on Cotton fibres (Cellulose) but inactive on methylated cellulose viz., CMC and Cx known as de-polymerise CMC

by random of terminal cleavage of B1, 4 glucoside linkages in cellulose polymers.

Mehta (1976), later studied the effect of phenolic substances on polygalacturonase (PG) production of *A. solani* and *A. tenuis* and observed decreased production of polygalacturonase. The degree of inhibition was also found varying with the type of compound and its concentration. The growth of pathogen was also recorded as inhibited by phenolics. Byrde *et al.* (1959); Patil and Dimond (1967) and Reddy and Mahadewan (1967), observed that phenolic compounds also inhibited the growth and synthesis of polygalacturonase (PG) in *A. solani* and *A. tenuis*.

Laxminarayana and Reddy (1978), studied the production of cellulase *in vivo* and *in vitro* and observed that cellulolytic activity of *A. tenuis*, *A. alternata*, *Helminthosporium hawaiiense*, *H. speciferum*, *Curvularia lunata*, *Hendersonula toruloidea* and *Phomopsis mangiferae* produced C1 and Cx enzymes and these fungi differed significantly, which is generally helpful in relation of pathogenic potentialities during infection process. In general C.M.C. supplemented media established as more conducive to enzymatic activity except *P. mangiferae*, which showed maximum activity on Asthana and Hawker's medium. None of the fungi showed C-1 activity.

Mehta and Mehta (1979), also studied the effect of nine carbohydrates and ten amino acids on the production of polygalacturonase transeliminase (PGTE) and pectinmethyl transeliminase (PMTE) of *Alternaria solani* and *A. tenuis* and found glucose to be as best carbohydrate source of these PGTE and PMTE in both the tested fungi. Starch was found in establishing only that carbohydrate inhibited the production of PGTE in *A. solani* and further observed that serine and histidine increased PGTE enzyme synthesis in *A. solani* being a substantial decrease in *A. tenuis* but no correlation between the fungal growth and the production/inhibition could be ascertained.

Prasad (1979), made studies on pectinolytic and cellulolytic activities of *Alternaria tenuis* (*A. alternata*) and found high on synthetic media. Wadie and Deshpande (1980) tabulated the effect of nitrogen sources, vitamins and trace elements on carboxymethyl cellulase *in vitro* by *A. tenuis* (*A. alternata*) causing. Leaf spot of cotton. Optimum conditions for the production of polygalacturonase and polymethylgalacturonase enzymes by a strain of *A. alternata* from Orange were established by Kunte and Shastri (1980).

Maximum amylase production by *A. humicola*, was observed after 8 days

inoculation (Singh and Bhatt, 1985).

Tak *et al.* (1985), observed the influence of pH and incubation period of enzyme produced by *A. alternata* causing Fruit rot of Apple, and found that polymethylgalacturonase activity was maximum in culture filtrates from glucose potassium nitrate medium with 1.0 per cent pectin after 9 days of incubation at pH 4.0 and 5.0, whereas polygalacturonase activity was maximum in pectinless medium after 9 days incubation at pH 6.0. They also recorded that cellulytic enzyme activity was maximum in a medium with 1.0 per cent after nine days at pH 6.0.

Stinson and Moreau (1986) and Hiltunen and Soderhall (1992), reported the presence of Alternarial-o-methyl transferase from *Alternaria alternata*. Dowd and Sheehan (1994), recorded the presence of glutathione-s-transferase from the fungus *Alternaria alternata*.

Dunaosku *et al.* (1995), studied the synthesis and secretion of extracellular protease enzymes and on the effect on conditions of cultivations by fungus, *Alternaria alternata* and *Fusarium oxysporum*.

NUTRITIONAL STUDIES

EFFECT OF CARBON SOURCESS ON THE GROWTH AND SPORULATION OF THE PATHOGEN

To find out the best carbon source for the growth and sporulation of *A. alternata*, several workers investigated the role of various carbon sources in the nutritional study of this pathogen and reported their findings.

Tandon and Grewal (1954), studied the nutritional requirements of *A. alternata* and observed that out of different carbon sources tested, sorbitol, glycerine, erytherifol, manmitol and dulcitol gave significantly good growth; glucose, galactose, mannose, maltose, lactose, sucrose, raffinose, dextrin and starch gave moderate growth, whereas poor growth was recorded in xylose, rhamnose, arabinose and inulin. This fungus failed to grow in absence of carbon source in the medium. The best sporulation of the fungus was recorded on xylose, glucose, galactose, mannose and lactose; fair on sucrose, raffinose and sorbitol, while poor sporulation was observed on glycerine, erytheritol, mannitol and dulcitol. However arabinose, maltose, starch, dextrin, inulin and the control were found as failed to induce the sporulation.

Grewal (1956), studied the carbon nutrition of *A. alternata* strain "B" and

found out of 19 sources the sugar alcohols supported good growth, whereas good sporulation was noticed on all the disaccharides. However best growth was obtained on sorbitol. No correlation was obtained between the growth and sporulation.

Tandon and Chaturvedi (1963), studied the carbon requirement of *A. alternata* and found that glucose closely followed by sucrose and maltose supported the best growth and sporulation of the fungus. Amongst pentoses, polysaccharides and alcohols, xylose, starch and sorbitol proved to be good source of carbon.

Singh and Khanna (1966), found mannitol to be the best source of carbon for the growth of *A. alternata*. Sucrose and maltose were also recorded to support good growth and sporulation of the pathogen.

Saad and Hagedorn (1970), observed that mannose, dextrose and maltose proved to be the best carbon source and fructose supported good growth.

Goyal (1977), found maximum growth of *A. alternata* on maltose followed by sucrose, starch, glucose and lactose though better sporulation was supported by sucrose.

Mathur and Sarbhoy (1977), done the nutritional studies on two isolates of *A. alternata* from Sugar beet and found sucrose to be the best source of carbon for both isolates.

Thind (1977), observed that galactose, glucose, fructose, mannose and sucrose proved to be good carbon sources for *A. alternata*. Fair growth was obtained on maltose and lactose, while poor on pentose sugars, starch and raffinose.

Gupta *et al.* (1979), observed *in vitro* tests that monosaccharides supported better mycelial growth and sporulation than disaccharides and polysaccharides. Sucrose supported poor growth but good sporulation.

Susuri and Hagedorn (1986), found that mannose, dextrine and fructose, were the best carbon sources for the growth of *A. alternata*.

Chattananavar *et al.* (1988), observed that a variation in utilization of sugar was evident in studies on *A. alternata* *in vitro*. According to them lactose

supported maximum growth of fungus.

Pandey and Verma (1992), found that the virulence of *A. alternata* to two cultivars of Pigeonpea (BDN-1 and TAT-10), were influenced by the pre inoculation nutritional status of the pathogen. *A. alternata*, was more virulent to BDN-1 following culture on media containing galactose, sucrose or maltose but was virulent to TAT-10, when grown on glucose and maltose.

EFFECT OF DIFFERENT SOURCES OF NITROGEN ON GROWTH AND SPORULATION OF THE PATHOGEN

Studies on nitrogen nutrition of *A. alternata*, have also been made by several workers.

Grewal (1956), recorded that growth of *A. alternata* was significantly improved by magnesium nitrate, calcium nitrate, ammonium acetate and ammonium oxalate, while other ammonium salts viz, ammonium sulphate and ammonium chloride were poor sources of nitrogen. No growth was observed with sodium or potassium nitrate or without only nitrogen source. The best sporulation was noticed on magnesium and calcium nitrate and peptone, fair or urea and reduction of growth in ammonium salts and being completely inhibited by ammonium chloride, ammonium sulphate and thiourea.

Tandon and Chaturvedi in 1963, observed that nitrates of potassium, sodium and calcium were good sources for the growth of *A. alternata*. Amongst organic nitrogenous sources glutamic acid, aspartic acid and phenylalanine supported good growth of the fungus but the fungus exhibited poor sporulation on glutamic acid and failed to sporulate on phenylalanine. Subsequently Singh and Khanna (1966), also observed good growth and sporulation of *A. alternata* on potassium nitrate. Aspartic acid also supported good growth of pathogen but induced poor sporulation.

Saad and Hagedorn (1970), observed in *vitro* studies that one of the isolates of *A. alternata* exhibited good growth of casein hydrolysate, glutamic acid, asparagine and tyrosine, while peptone and sodium carbonate supported excellent growth of the other isolates.

Goyal (1977), found that *A. alternata* utilized nitrate nitrogen more efficiently than ammonium salts except ammonium oxalate, which gave good response. Good sporulation was observed on sodium nitrate followed by potassium nitrate and calcium nitrate. Ammonium salts did not exhibit any

sporulation except ammonium nitrate, which supported poor sporulation.

Susuri and Hagedorn (1986), found peptone to be the good source of nitrogen for the growth of *A. alternata*.

Chattananavar *et al.* (1988), recorded that a variation in utilization of nitrate sources was apparent in studies on *A. alternata* *in vitro*. Sodium nitrate and arginine, were the best inorganic and organic sources of nitrogen. No growth of the fungus was observed on citric acid and ammonium nitrate.

Pande and Verma (1992), concluded that *A. alternata*, preferred ammonium salts. This fungus produced moderately good growth and sporulation on ammonium chloride but could infect hardly six leaves in cultivar BDN-1 and nine leaves in TAT-10 of *Cajanus cajan*, L.

SUSCEPTIBLE GROWTH PERIOD OF THE HOST :-

Avoidance of pathogen is one of the basic factor for management of plant diseases. Majority of diseases can be eradicated by selection of change of land and alteration in time of sowing being an aim of measures. The aim of management of disease is to enable the hosts to avoid contact with pathogen or the susceptible stage of plant and favourable conditions for pathogen should not coincide. In most of the diseases the disease incidence is most severe, when the susceptible stage of plant and favourable conditions for development of pathogen coincide. At the time of sowing the main criterion is required to be given main importance that all the conditions viz; susceptible stage of plant growth and climatic conditions favourable for maximum activity of pathogen do not fall at the same time.

Older plants of Bean, were observed as most susceptible to attack of *Alternaria alternata* than younger ones (Saad and Hagedorn, 1969 and Crissan *et al.* 1979).

Kamal *et al.* (1971), observed that in Cotton cultivars the optimum age of leaves for infection of *Alternaria alternata*, was 30-45 days and also observed to cause inhibition of spore germination and germ tube growth by juice of seedlings and leaves of resistant cultivars.

Stavely and Slana (1971), found that development of pustules due to *Alternaria alternata* on young leaves of *Nicotiana tobaccum*, was less than 1.0 mm. in comparison to older leaves, which were more than 8.0 mm. in diameter. It determines that older leaves were more susceptible than younger leaves.

Mehta *et al.* (1975), also studied the pronounced effect of age of host and pathogen development of disease caused by *Alternaria solani* and *A. tenuis*.

Sarkar and Sengupta (1978), reported that 20 days old plants of Mustard were resistant to infection incited by *Alternaria brassicicola* in comparison to 30 days old plants, which were slightly infected and susceptibility increased with increase in age of plants. Further Chahal (1986), studied the susceptibility of Brown Sarson to *Alternaria brassicae* increased with the increase in age of plants.

The young plants of *Lycopersicon esculentum* exhibited high degree of resistance to early blight caused by *Alternaria solani*, while with increase in age of plants caused loss in intensity of resistance. The maximum incidence of disease was obtained in 4 months old plants and minimum in 20 days old plants (Bedi and Dhiman, 1980).

The older leaves of *Helianthus annuus*, L., were found susceptible to *Alternaria helianthi* (Allen *et al.* 1983 and Godnoy and Fernandes, 1985). Contrary to these findings the younger plants of Sunflower were found equally susceptible to *Alternaria helianthi* (Jeffrey *et al.* 1984).

Droby *et al.* (1984), recorded that young plants of Potato at the 10-12 leaf stage were less susceptible than adult plants against *A. alternata*.

Suhag *et al.* (1985), found that 30-35 days old leaves of *Raphanus sativus*, were more susceptible to *Alternaria alternata* and require less incubation period for inducing gradual multiplication of disease with increase in age of plants in field especially the lower leaves, which are older having higher relative humidity than upper and younger leaves.

Singh and Shukla (1986), observed maximum disease due to infection of *Alternaria alternata* on 4 months aged plants of *Solanum melongena* followed by 135 and 105 days old plants and on the basis of these findings, it was determined that older plants were found more susceptible to disease than younger ones.

Shukla (1987), reported that two months old plants of triticales were more susceptible to infection of *Alternaria alternata* in comparison to 30 and 45 days aged plants.

Singh *et al.* (1987), studied the effect of date of planting, crop age and

climatic factors on Potato leaf spots and observed that crops sown on 20th October developed least epidemics of disease caused by *Alternaria alternata* and *Phoma* sp. and gave highest yield, crop age and relative humidity were found positively correlated with disease incidence, whereas maximum temperature was found negatively correlated. The total effect of environmental factors and incidence of disease was found significant. The crop age was found alone to regulate the disease. The susceptibility to leaf spots of mature plants was regulated by an increase in soluble sugars and decrease in total nitrogen content.

Ghemawat *et al.* (1989), observed that artificial inoculation of 5 days aged plants of susceptible *Helianthus annuus*, L. variety EC-68414 infected with *Alternaria helianthi* resulted in 100.0 per cent mortality, which was found decreased to 13-30 per cent, when one month old plants were inoculated. Two months old plants expressed no mortality after inoculation and sustained only moderate loss on seed and oil yield.

EPIDEMIOLOGY OF DISEASE -

Epidemiology deals the effect of temperature, humidity, wind velocity and rain fall affecting incidence and disease severity. It is necessary for forecasting and planning of strategy for effecting control against a disease. Dey (1933), studied the effect of humid condition on the prevalence of disease of *Alternaria* blight disease of Linseed and noticed the severity of disease in undrained fields, where air round the plant, was found as wet for a long duration.

Arya and Prasad (1952), recorded cent per cent Linseed blight disease development at the temperature range of 26°C - 30°C in a saturated atmosphere for 72 hours after inoculation. High humidity and temperature at a range between 12.6°C - 18.30°C favoured prevalence of development of Leaf spot disease of Crucifers caused by *Alternaria brassicae* (Dey, 1948; Mc-fariane *et al.* 1984; Louvet 1958; Loof 1959 and Louvet and Billotte (1964).

Domach (1957), recorded that persistent relative humidity of 95.0-100.0 per cent for at least 18 hours was essential at the temperature range of 21°C and 27°C for at least three successive days. Ashour and El-Kadi (1958), was of opinion that optimum temperature 25°C and 75.50 per cent relative humidity was essential for best growth of *Alternaria tenuis*, while optimum temperature of 27°C was required for germination of spores and infection as reported by Kamal *et al.* (1971).

Prabhu and Prasad (1966), recorded the maximum infection of *A. triticina* at a temperature range of 20°C - 25°C. They also observed that the optimum temperature for infection was somewhat lost to that required for optimum growth of pathogen in culture.

Bakos *et al.* (1967), studied the incidence related to rain fall. The epidemiology of Leaf spot of Bean incited by *A. tenuis*, was studied by Saad and Hagedorn (1969), who found that relative humidity over 95.0 per cent was found to be essential for initiation of disease and further found that disease development was minimum at 28°C and severe at 16°C.

Pamubov (1972), studied the epidemiology of Brown spot of Virginia Tobacco caused by *A. alternata* and found favour of infection at temperature range of 20°C - 21°C, high rain fall and relative humidity more than 67.0 per cent favoured infection. The effect of ecological factors on sporulation of *Alternaria helianthi* and infection to Sunflower plants was studied by Acimovic (1974).

Rao and Raj Gopalan (1982), observed good growth and sporulation at 29°C - 31°C. Sarkar and Sen Gupta (1978), studied some aspects of epidemiology of Alternaria blight of Mustard caused by *A. brassicicola* and observed that optimum temperature for growth of mycelium was 27°C. 65.0-100.0 per cent relative humidity favoured the good mycelial growth. They also observed that at least 24 hours of saturated atmosphere, was required for proper development of disease.

A study on the effects of temperature, relative humidity, age of pathogen and age of fruits of *Lycopersicon esculentum*, was made by Mehta *et al.* (1975) and observed that temperature of 28°C and 100.0 per cent relative humidity were more effective in disease development. Semi-ripe fruits were found to be more susceptible in comparison to ripe fruits.

Successful infection caused by *Alternaria triticina*, was also observed in 48 hours in a saturated atmosphere in comparison with 73-96 hours for a similar degree of infection caused by *Alternaria alternata*. The establishment of maximum lesion development by the pathogen was also recorded at 10°C and greater intensity of development was recorded at 10°C and greater intensity of development was recorded greater at 28°C than at 15°C or 30°C (Ram and Joshi, 1978).

Sarkar and Sen Gupta (1978), studied some aspects of epidemiology of Alternaria leaf spot of Mustard caused by *A. brassicicola* and reported optimum temperature for mycelial growth was 27°C. Good growth occurred at relative humidity of 65.0 - 100.0 per cent. They also observed that at least 24 hours of saturated atmosphere was required for proper development of disease. Rao and Raj Gopalan (1979), observed good growth and sporulation at 29°C - 31°C.

Prasad and Roy (1979), found that pathogens viz., *Alternaria alternata* and *Chochilobolus spicifer* caused pronounced rotting of *Solanum melongena* and Sponge gourd at 26°C and 90.0-100.0 per cent relative humidity. Dickson and Bottomley (1980), studied the effect of relative humidity, temperature and nutrient conditions on germination and growth of *Alternaria alternata* and *Cladosporium* sp.

The germination of conidia of *A. helianthi* causing leaf spot of Sunflower was induced and favoured by a temperature range of 25°C-28°C in presence of free water on the surface of leaf. The occurrence of maximum infection was observed if subjected to continuous 12 hours period of leaf wetness (Allen *et al.* 1983).

The development of leaf spot disease caused by *A. alternata*, was favoured if subjected to relative humidity, frequent irrigation and high rate of application of nitrogen as reported by Chattanavar *et al.* (1984).

The leaf spot disease of *Solanum melongena*, was favoured by the climatic factors ie. temperature and relative humidity ranging between 24°C - 26°C and 47.30 to 51.20 per cent relative humidity respectively. Scanty rains and long dry spell had more adverse effect on development of disease than intermittent rainfall. The infection rate was not found uniform and disease was increasing with increasing rate of 0.323 per cent unit per day (Dingar and Singh, 1985).

The correlation between the climatic conditions with the development of disease of Pod and leaf blight of *Raphanus sativus* seed crop caused by *Alternaria alternata*, was studied by Suhag *et al.* (1985), who found that conidial dispersal was not influenced by daily temperature fluctuations but have a positive relation with rain fall and relative humidity. There was considerable increase in the disease, when mean temperature and relative humidity were 20.60°C - 23.8°C and 57.90-67.40 per cent respectively.

Ghewande (1986), correlated the temperature between 25°C - 29°C and

87.0 per cent relative humidity was most suitable for development of *Alternaria* leaf spot of *Arachis hypogea* caused by *Alternaria alternata*, whereas Ungaro and Azavedo (1986), found good sporulation in dark at $27\pm 1^{\circ}\text{C}$.

Bhargava and Khare (1988), studied the epidemiology of *Alternaria* blight of Chickpea (*Cicer arietinum*, L.) incited by *A. alternata* and observed the appearance of disease first at flowering stage when temperature ranged between 25°C - 27°C with relative humidity of around 80.0 per cent in weather conditions of season specifically periodic rains. Consequently high relative humidity favoured disease development. The severity of *Alternaria* leaf spot of Cotton was studied by Hiremath *et al.* (1988) for six years in Karnataka and its correlation with climatic factors. He observed slow and fast development of disease during August-September (Vegetative period) and November (Peak boll development stage) respectively. Rains 10 mm. or more commenced in November favoured quick development of disease.

A positive correlation between disease severity and relative humidity causing *Alternaria* blight of *Helianthus annuus* L. caused by *Alternaria helianthi*, it was found that temperature has a significant importance towards its infection. The yield was found 24.0-46.0 per cent and 29.0-35.0 per cent germination was recorded (Hiremath *et al.* 1990).

Gupta and Pathak (1990), observed the optimum temperature for Fruit rot of Papaya (*A. alternata*) was 25°C . Severity increased with an increase in relative humidity, reaching maximum at 10.0 per cent.

Patel and Patel (1991), reported that temperature of 25°C - 40°C and high relative humidity favoured the development of Tomato rots in open market caused by *A. alternata*.

Infection due to *Alternaria alternata*, was found favoured moderately at a normal temperature, normal relative humidity and dry rainless days (Kumar, 1992).

Awasthi and Kolte (1994), observed that relative humidity (76.70 per cent), total rainy days (76), rain fall (7.70 mm.) and minimum temperature ($7-10^{\circ}\text{C}$) increased *Alternaria* blight severity on leaves of Rapseed and Mustard. Relative humidity and total rainy days, were significant contributors to the spread of pod infection.

Borker and Patil (1995), reported that weather parameters with the

disease development of Alternaria leaf spot of Sunflower caused by *Alternaria helianthi* were temperature of 25.90°C - 33.7°C with relative humidity of 89.0 - 95.0 per cent. Disease development was higher, when sufficient intermittent rain/showers (1.20 to 12.80 mm. in a meteorological week), were received during progress of disease, however less rain or heavy rains did not caused much disease as compared to optimum intermittent rains.

EFFECT OF PATHOGEN ON CERTAIN BIOCHEMICAL CONSTITUENTS OF DISEASED PARTS OF DOLICHOS BEAN (*Dolichos lablab*, L.)

The biochemical resistance depends upon the pre-existing or performed substances in the host or it may also be induced through substances induced by pathogen after exciting the infection of host plants.

Horsfall and Dimond (1957), was credited to differentiate the diseases on the basis of amount of sugar content present in the host plant into two categories viz., "high sugar" and "low sugar" diseases. Under the category of low sugar disease, Ghemamat and Prasad (1972), studied Alternaria blight (*A. burnsii*). Aulakh and Grover (1970), recorded reduction in intensity of infection after getting incited with *A. alternata* causing diseases of fruits of *Lycopersicon esculentum*.

Diffusion of fungal metabolites were found in alternation of leaves of *Nicotiana tabaccum* infected with *A. alternata* resulting decrease in plastid pigments and reducing sugars (Main, 1971).

Bhatia *et al.* (1972), found total tannin and phenols in the resistant varieties of *Lycopersicon esculentum* to *A. solani*, were higher than susceptible varieties, while sugar and total carbohydrate (Sugar starch) found decreased in all the diseased leaves of Potato caused by *A. solani*.

Chopra and Jhooty (1974), studied the biochemical changes in resistant and susceptible varieties of Water-melon due to infection of *Alternaria cucumerina* and found no significant changes in the glucose content. Sucrose, raffinose and lactose decreased progressively towards the necrotic spot. Later Chopra *et al.* (1974), recorded that after infection by *A. cucumerina* the resistant varieties were rich in phenolic compounds in comparison to susceptible varieties and determined that phenolic compounds alone or in combination with amino acid may play important role in the mechanism of disease resistance.

Kumar (1974), reported that high percentage of wax content in the variety

of *Triticum vulgare*, were resistant to infection of *A. triticina*, low in susceptible varieties and intermediate in moderately susceptible varieties. Later amount of sugar and starch was found low in both resistant and susceptible varieties of wheat against *A. triticina* by Kumar and Rao (1980), while total nitrogen was found increased in susceptible and decreased in resistant varieties. Phenolic contents, were also found increased in leaves of resistant varieties, which were found decreased in susceptible varieties.

The biochemical changes of varieties of Cotton resistant and susceptible to *Alternaria macrospora*, were studied by Bhaskaran *et al.* (1975), who reported that polyphenol oxidase and peroxidase activities were higher in resistant varieties but in inoculated varieties with the pathogen peroxidase activity was found increased and polyphenol oxidase activity was decreased. Total amino-nitrogen were high in susceptible varieties, which were found increased in susceptible and decreased in resistant varieties. Phenolic compounds were also found increased in leaves of resistant varieties, which were found decreased in susceptible varieties.

The biochemical changes of varieties of Cotton resistant and susceptible to *Alternaria macrospora*, were studied by Bhaskaran *et al.* (1975), who reported that polyphenol oxidase and peroxidase activities were higher in resistant varieties but in inoculated varieties with the pathogen peroxidase activity was found increased and polyphenol oxidase activity was decreased. Total amino-nitrogen were high in susceptible varieties, which were found increased after inoculation. Further Padmanabhan and Naryanaswamy (1978), observed that *A. macrospora* infected leaves of Cotton exhibited a reduction in Chlorophyll content.

Bhaskaran and Kandaswamy (1977), reported that respiratory activity in halo and pre-halo areas was decreased in the necrotic tissues of leaf spot of Sunflower caused by *A. helianthi*. In halo-zone areas the catalase activity was also found high and low in necrotic tissues without any significant alteration in pre-halo zone. The rate of photosynthesis, chlorophyll contents, carotene, xanthophyll and starch were found in all the three zones, whereas β -amylase activity increased. Total soluble sugars, glucose and fructose were found decreased in necrotic and halo areas and without any significant change in sugar content in pre-halo tissues. In necrotic regions sucrose level was found low and high in halo regions.

Agrawal and Bisen (1978), observed the difference in phenolic fraction of

resistant and susceptible varieties of Tomato prior and after inoculation with *A. alternata*. They recorded that compounds, were primarily accumulated during lesion development and to a greater extent in resistant varieties.

Ali and Roy (1981), studied the young Carrot leaves infected by *A. dauci*, were having higher content of reducing sugars and tannin and low content of nitrogen and protein, which induce resistance in the leaves against disease. Dixit and Gupta (1983), found decline in disease development due to presence of sugar in leaves of *Hordeum vulgare* infected by *A. alternata*.

Higher amount of sugar and phenols in young leaves of Brown Sarson infected with *A. brassicae* account of development of greater resistance in comparison to older leaves.

Total nitrogen, phosphorus and potash contents, were recorded as decreased in blighted, halo and healthy tissues of Onion leaf in close association to diseased tissue caused by *A. porri* as compared to the healthy varieties on Onion (Khatri *et al.* 1986).

In resistant varieties of *Sesamum indicum*, L. against, *A. sesami* the wax, phenol and chlorophyll contents were found higher than the susceptible varieties (Gupta *et al.* 1987).

Bhargava and Khare (1988), observed that leaves of resistant cultivars of *Cicer arietinum*, L. against Alternaria blight caused by *A. alternata* induced the presence of phenols in rich percentage, orthodihydric phenols, sugars, nitrogen, phosphorus and potash as compared to susceptible varieties.

SURVIVAL, PERPETUATION AND SPREAD OF THE PATHOGEN

A very little information is known about survival, perpetuation and spread of leaf spot of Dolichos bean caused by *Alternaria alternata*.

Higgins (1947); Wheeler (1958); Raman and Lucas (1963) and Mawana (1983), studied the mode of infection, survival and spread of different diseases caused by *Alternaria* sp. and observed that development of disease in next season takes place through seed borne pathogens through plant debris, but unable to survive in soil in the form of mycelium or conidia.

Kapoor and Hingorani (1958), during the course of their studies on the role of seeds in initiation of the disease observed that unsterilized seeds of *Solanum melongena*, obtained from infected plants, when sown in sterilized soil only 25.0

per cent seeds germinated and rest ungerminated seeds were found rotted due to infection of *Alternaria* similar to Brinjal isolate. Cotyledons of germinated seeds were severely blighted and the seedlings that emerged from them were destroyed so the pathogen was considered by them to be seed transmissible.

Prabhu and Prasad (1967) and Bhowmik (1969), isolated *Alternaria triticina* and *A. alternata* from wheat seeds having brown to black discolouration. *A. alternata*, was isolated from the seeds of other crop viz., Maize, Oat, Barley and Sugarbeet (Radulescu and Ngru, 1965), Pigeonpea (Kumar and Patnaik, 1985) etc.

The infested soil and plant debris, were found by Gamamat and Prasad (1972) to spread disease from one crop season to another by perpetuation of *A. burnsii* causing leaf blight of Cumin. Further they discussed that infected seeds play an important role in perpetuation of pathogen and primary infection. The secondary infection was performed through aerial infection. Singh and Bedi (1980), also described the penetration and infection process of *Alternaria alternata*.

Kumar and Neema (1973), studied the mode of perpetuation and recurrence of leaf blight of wheat and observed that during non- crop season, *A. triticina*, survived as conidia on the seed surface as well as dormant mycelium inside the seed coat but the viability of the fungus was lost, when infected plant debris left over in the field was exposed to summer temperature.

The seeds infected by *Alternaria alternata* formed on affected head of Sunflower, were found capable in perpetuation and transmission of disease by Agrawal and Singh (1974); Narain and Saksena (1981); Prasad and Singh (1983); Vijayalakshmi and Roy (1985) and Krishnappa and Shetty (1990).

Shukla *et al.* (1978), reported the leaf blight of Wheat caused by *Alternaria alternata*, was isolated from the underlying soil and the plants grown in pods containing contaminated soil showed disease symptoms.

Alternaria macrospora, causing leaf spot of Cotton was found to be seed borne in 90.0-100.0 per cent of infected bolls (Padaganur, 1979). Spores adhering on the seed coats in cent per cent of seeds collected from infected bolls and 50.0 per cent of these infected seeds, were observed as primary source of infection for perennation of disease.

Susuri *et al.* (1980) became successful in reducing diseased healthy plants

after inoculation with spore suspension of *A. alternata* obtained from infected Pea plants and they also investigated the mode of penetration leaf tissues.

Zazzerini and Buonauro (1981), reported *Alternaria alternata* and *A. carthami* on infected leaves of *Helianthus annuus*, L. and found that infection takes place through seeds and favoured by rain fall during plant growth.

Alternaria helianthi, causing leaf blight of *Helianthus annuus*, was established as a seed borne in nature having ability to overwinter in diseased plant refuse from year to year (Herr and Lipps, 1982 and Allen *et al.* 1983).

Singh and Suhag (1983), recorded that the pathogen, *Alternaria alternata* isolated from *Raphanus sativus* causing leaf and pod blight remained viable in seeds obtained from diseased pods as well as debris for a longer period to infect succeeding crop. The prevalence of seed borne infection of *Alternaria alternata* has also been reported for the spread of disease in secondary spread of pathogen took place through air borne Pigeonpea by conidia produced on infected leaves. Kumar and Patnaik (1985).

The effect of plant age on susceptibility and seed infection was studied by Godnoy and Fernandes (1985) and found plants to be more susceptible on older leaves in some plants. Incidence was not found reduced by seed treatment with mercuric chloride or sodium hypochlorite.

The plant parts and seeds of Sunflower infected by *Alternaria helianthi* showed the soil and seed borne nature of the disease as observed by Jeffery *et al.* (1984) and Raut (1985).

Singh (1987), studied the survival and perpetuation of *Alternaria alternata* causing leaf spot and fruit rot of Brinjal and found that the pathogen survived in plant debris up to one year, when kept at room temperature ranging between 25°C- 28°C, under laboratory conditions but was able to survive up to six months subject to buried in soil. Infested soil, plant debris and seed served as a source of primary infection. Debris was kept on or above ground the pathogen remained viable for eight months under both field and laboratory conditions but viability gradually decreased in all the conditions.

Prasad and Reddy (1986), found that the pathogen, *Alternaria alternata* was able to survive for three months causing alternariosis of *Arachis hypogaea* falling on soil surface and exposed to natural conditions and up to eight months on leaves in laboratory.

Alternaria alternata causing excessive leaf spot of *Vicia faba* in experimental plots was isolated by Simay (1987), who found successful inoculation with conidia in glasshouse. Symptoms being most marked on plants previously infected by *Aphis fabae* or *Tetranychus urticae*, were present. He observed seed transmission inspite of sterilization of seed surface.

Secondary spread of pathogen took place through air borne conidia developed on infected leaves. Sultana *et al.* (1980) also observed the existence of seed borne nature of *Alternaria alternata* causing Fruit rot of Chillies.

Gupta and Pathak (1988), found *Alternaria porri* causing Purple Blotch of Onion survived in diseased Onion leaf and seed stalk debris burried in soil at 5.0 and 7.5 cm. respectively for 12 months. Thus it is established that pathogen survived in infected plant debris in the soil and in infected bulbs.

Ansari *et al.* (1989), made the studies on the mode of survival and perpetuation of *Alternaria brassicae*, attacking Rape seed and Mustard plants. The pathogen was found viable in diseased plants debris and seeds of infected plants serving as a primary source of inoculum. The pathogen in its nature was found internally seed barne and affected seeds caused Damping off of seedlings. Air borne conidia of aerial parts of infected plants were established for secondary spread of disease.

Bashon *et al.* (1991), studied the wind dispersal of *Alternaria alternata* causing leaf blight of Cotton.

Anwar *et al.* (1994), found that percent infection and disease incidence of Pea caused by *A. alternata* ranged from 20.0-70.0 per cent and 1.0-6.0 per cent respectively. Infection decreased seed germination potential.

VARIETAL SCREENING -

Use of resistant varieties is the most effective, simple, practical and economical method for the control of epidemics of disease, which not only ensure protection but also save energy and money spent on the other measures employed for control of disease. The resistant varieties are significant due to the fact that it gives advantages to the farmers to provide more benefits other than the investments in need and other input as well as output if pathogen co-operates and resistance persists (Singh, 1984). No work appears to have been done on the source of resistance to Dolichos bean against *Alternaria* leaf spot caused by *Alternaria alternata*. Some work has been done on varietal resistance

against *A. alternata* in other Beans.

Gurha *et al.* (1981), reported that out of 39 varieties of Broad bean screened under controlled conditions, the large podded, bold seeded, V-Japan and V-Japan Yanatwase from Japan and the profusely fruiting Dholi-3 from India proved resistant to *A. alternata*.

Issa and Oliveira (1986), tested 7 varieties and 3 varietal mixtures of French bean obtained by crossing for horizontal resistance and found that Marina-80 was least affected by *A. alternata* pod lesions, whereas Sefe-3 had the lowest leaf lesion. Khalil *et al.* (1986), observed that ILB-938 varieties of Faba bean (*Vicia faba*, L.) had the best resistance against *A. alternata* followed by ILB-592.

HOST RANGE STUDIES -

Alternaria tenuis (*A. alternata*) is common polyphagous cosmopolitan saprophyte as well as facultative parasite parasitizing on many plants eg. *Avena sativa* (Oat), *Abutilon* sp., *Abelmoschus esculentus* (Bhindi), *Althea rosea* (Holyhock), *Arachis hypogea* (Groundnut), *Brassica juncea* (Rye), *B. campestris* (Mustard), *B. oleracea* (Cauliflower), *Carthamus tinctorius* (Safflower), *Chenopodium album* (Bathua), *Chrysanthemum maximum* (Guldawadi), *Cucurbita maxima* (Pumpkin), *Cucumis melo* (Muskmelon), *Cucurbita sativus* (Kheera), *Citrullus fistulosus* (Tinda), *Cicer arietinum* (Chick pea), *Cajanus cajan* (Pigeon pea), *Corchorus capsularis* (Sunn-hemp), *Cynodon dactylon* (Doob grass), *Dahlia* sp., *Euphorbia nerifolia* (Singhara), *Gossypium* sp. *Glycine max* (Soyabean), *Hibiscus rosa-sinensis* (Gurhal), *Hordeum vulgare* (Barley), *Lens esculenta* (Lentil), *Lagenaria vulgaris* (Bottle gourd), *Momordica charantia* (Karela), *Oryza sativa* (Paddy), *Pennisetum typhoides* (Bajra), *Pisum sativum* (Pea), *Phaseolus radiatus* (Urd), *Ricinus communis* (Castor), *Saccharum officinarum* (Sugar cane), *Sorghum vulgare* (Jowar), *Sida acuta*, *Spinacea oleracea* (Spinach), *Triticum aestivum* (Wheat), *Trifolium alexandrinum* (Dersoom) and *Zea mays* (Maize) etc. Being weak in nature the fungus is found associated with or producing "Damping off of seedlings in Tobacco", "Black point of wheat", "leaf spots", "leaf blights" and various other diseases. It also inhibits seed germination (Groves and Skolko 1944; Neergaard, 1945; Siddiqui, 1963 and Rao, 1969 and 1971).

Different infection experiments of *Alternaria* sp. carried out by Young (1926) against various hosts suggested that *A. tenuis* may appear as a weak

parasite on many plants and found in number of hosts viz., *Brassica oleracea*, *B. rapa*, *Capsicum annuum*, *Daucus carota*, *Hordeum vulgare*, *Solanum nigrum*, *Spinacia oleracea*, *Zea mays* etc., as true hosts of *A. tenuis*.

Neergaard (1945), characterized *A. alternata* as a pronounced polyphagous facultative parasite. In order to determine the host range of this species, mono spore inoculation experiments were carried out on 36 plants species belonging to 14 families, ten principal ones being Wheat, Onion, Carnation, Cabbage, Peas, godetia hybrids, Cucumber, Carrot, Tomato and lettuce. Positive results were obtained in inoculation tests on Onion, Carrot, Carnation, Peas and Wheat.

Kapoor and Hingorani (1958), in host range studies of *A. alternata* included *Brassica oleracea* L., *Lycopersicon esculentum* L. Kurst., *Nicotiana tabacum*, L., *Solanum nigrum* L. and *S. tuberosum*. They found that it infected only *Solanum tuberosum* L. and *Lycopersicon esculentum*, L. Kurst.

Singh and Tandon (1967) reported that the isolation of *A. alternata* from Mango infected the leaves of *Solanum melongena*.

A survey made by Mehrotra and Narain (1969), of *Alternaria* disease against 55 various hosts belonging to 25 families, infected by various species of *Alternaria*. Twenty hosts were found as a new record from world and 28 were new hosts from India. Similarly Narain and Saksena (1974, 1975 and 1978) also studied the different species of *Alternaria* occurring on various hosts a new record from India.

Nine species of *Alternaria* from ten vegetables, were isolated by Crisan (1976). Out of which *A. alternata*, was the most common pathogen attacking nine vegetable crops yielding plants including Chillies, Tomato, Bean and Egg plant in various degrees. Isolates of *A. alternata*, obtained from Tomato and Egg plant were found more prevalent in comparison of Bean plants.

The host range and virulence of isolates of *A. tenuis* (*A. alternata*), *A. tenuissima* and *A. triticola*, out of 27 plant species were studied by Patil *et al.* (1981), who further studied the host range of *A. alternata* and *A. solani* causing Early blight of *Solanum tuberosum* and found wider host range of *A. alternata* than *A. solani*, while Narayanappa (1982), recorded wide host range isolated from *Raphanus sativus*.

Singh and Suhag (1983), reported *Alternaria alternata* by isolating them from *Raphanus sativus*, pathogenic on Cabbage, Cauliflower, Spinach, Tomato,

Convolvulus arvensis and *Trienathema monocyna*.

In addition to various reports on the host range studies as mentioned above the *Alternaria alternata*, was implicated to cause leaf blight, leaf spot, Stem blight and Fruit rot on different field and ornamental plants eg. Castor (Tropova, 1930), Brinjal (Kapoor and Hingorani, 1958), Hollyhock (Sharma, 1958), Tomato (Tandon and Chaturvedi, 1965), Grape (Ratnam and Nema, 1967), Clusterbean (Chand and Verma, 1968), Makhana (Prasad and Haider, 1968), Chrysanthemum (Waterworth and Pavish, 1968), Sugar beet (Mukhopadhyay, 1968), Barley (Dhanraj, 1970), Chilli (Sauz and Hermilla, 1970), Singh *et al.*, 1984 and Singh (1987), Mung bean (Gupta, 1970), Tobacco (Stavely and Slana, 1971), Datura (Janardhan and Hussain, 1972), Wheat (Sokhi *et al.* 1972), Chillies (Sreekantiah *et al.*, 1973), Rose (Sezgin *et al.*, 1973), Chick pea (Shukla and Bhargava, 1976), Bajra (Gaikwad and Rane, 1977), Sunflower (Narain and Saksena, 1973 and 1981), Pear (Koul and Narain, 1981), Kodon (Gupta *et al.* 1982), Pigeon pea (Mehta and Sinha, 1982), Pea (Sharma, 1983), Sesame (Rani *et al.* 1983), Coriander (Prasad, 1983 and Khan *et al.* 1984), Cotton (Singh *et al.* 1984), Watermellon (Narain *et al.* 1985 and Mango (Prakash and Roof, 1985), Kondon (Gupta *et al.* 1987).

Narain *et al.* (1987), also reported the occurrence of two foliar diseases of Groundnut caused by *A. alternata* as new record of U.P.

CHEMICAL CONTROL -

The peculiar leaf spot of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* could not get the proper attention of Plant pathologists due to non-availability of useful devices related with control and other aspects. At present in agriculture either prophylactic treatment or by direct treatment against pathogens is one of the most important inputs in increasing and sustaining productivity. Use of chemicals is a practical device of controlling the diseases being the main method of control. The techniques employed in application of fungicides and its efficacy depends upon the nature of host and host parasite relationship, cultural practices employed to the crop and other economic aspects of the crop. A brief of resume of work done on bioassay of seed treatment and fungicides is given below.

(A) BIOASSAY TEST

Misra and Singh (1965), studied the toxicity and physical properties and

biological spectrum of certain copper and organic fungicides with *A. alternata* as test organism and found that Captan was able to inhibit mycelial growth of fungus.

Sahni and Singh (1967), observed the growth of *Alternaria alternata* in *vitro* as completely inhibited by treatment with Arasan, Thiram and Zineb. The best control of the disease was achieved with Captan, Ferbam and Maneb at 0.50-0.25% under laboratory conditions (Crisan *et al.*, 1970).

Misra and Singh (1970), also reported Captan as best fungicide to control the mycelial growth of *Alternaria tenuis* selected from copper and organic fungicides. Ram Krishan *et al.* (1971), found 0.15 per cent Dithane Z-78 and 0.20 per cent Duter as most effective.

Singh and Milne (1974), tested fifteen fungitoxicants under laboratory against *A. alternata* and Captafol, Dithane M-45 and Thiram were found to be most satisfactory.

The efficacy of 17 fungicides, was also studied on the spores of *Alternaria alternata* on Cotton by Youseff *et al.* (1977). Nine fungicides were tested under laboratory conditions against spore germination and growth of *Alternaria alternata* by Mathur and Sarbhoy (1977) and found that Aureofungin and Brestanol, were the most effective ones.

Thiram was concluded to be best fungicide by Kuthbutheen and Pugh (1978), who observed various degrees in growth rate and spore germination of *Alternaria alternata* causing Leaf spot *Glycine max* followed by Captan and Verdesan. Narain (1978), in laboratory studies recorded complete inhibition of the growth of *Alternaria alternata* in *vitro* by Difolatan, Plantav, Benlate, Bavistin, Dithane M-45, Dithane Z-78 and Duter but Crisan *et al.* (1979) found Vitavax, Acricide and Quinolate at 0.10 per cent and 0.20 per cent as effective against *A. alternata*, isolated from Bean. Utikar *et al.* (1979), reported Zineb and Dithane M-45 (Mancozeb) as more superior to other fungicides examined against the fungus, *A. alternata* isolated from Onion causing complete inhibition of all concentrations.

Ziram was found most suitable fungicide in *vitro* against, *Alternaria helianthi* by Bhaskaran and Kandaswamy (1977) and Mukewar and Gera (1980). Kolte *et al.* (1979), reported and proved Mancozeb dithiocarbamate to be most effective in control of disease up to 17.0 per cent - 33.0 per cent and increase in yield up to 43.0-65.0 per cent.

Desai and Patil (1982), tested the efficacy of eleven fungicides against *Alternaria macrospora* causing leaf spot of Cotton in *vitro* and found that RH-216 (Penapalin) and Pancotine (Quazatine) as most effective followed by Cuman-L (Ziram), Dithane M-45 (Mancozeb), Duter (Fintin hydroxide) and Blittox (Copper oxychloride).

Kalra and Sohi (1984) observed that except Calixin, Systemic fungicides were not effective against *A. tenuis* (*A. alternata*) in *vitro*. They further recorded that Thiram (0.05-0.20 per cent), Dithane M-45 (0.10-0.20 per cent) and Difolatan (Captafol 0.20 per cent), completely controlled the growth and other fungicides reduced it. It was also recorded that Dithane M-45 (Mancozeb) and Pancotin were as most inhibitory to growth of *A. macrospora* isolated from Cotton (Siddaramiah and Desai, 1984).

Ungaro and Azevedo (1984), reported Zineb (0.20 per cent) as most effective fungicide in producing morphological changes and control of the fungus against *A. alternata* and *A. helianthi* followed by Captan (0.25 per cent) in *vitro*.

Mali and Joi (1985), found Difolatan (Captafol), Thiram and Vitavax (Carboxin) as most effective against colony growth and sporulation of *Alternaria alternata*.

Sharma and Chauhan (1983), recorded that spore germination of *Alternaria macrospora*, *Curvularia lunata*, *Helminthosporium speciferum* and *Myrothecium roridum* isolated from Cotton was completely controlled by Blitox (Copper oxychloride), Dithane Z-78 (Zineb) and Difolatan (Captafol) at 100 and 500 ppm. and Dithane M-45 and Syllit (Dodine) at 500 ppm. The toxicity, physical properties and biological spectrum of certain copper organic fungicides was studied by Misra and Singh (1985), against *Alternaria tenuis* as test organism and found Captan to inhibit mycelial growth to a great extent.

Vasu and Rao (1985), recorded that, 2, 4-5 trichlorophenyl phosphite proved most effective against *Alternaria alternata*.

Vakalounak's and Malathrakis (1988), tested in *vitro* efficacy of 25 fungitoxicants, out of which Chlorothalonil, Dichlorflunaid, Guazatine, Iprodione and Maneb gave the best response against *A. alternata* of Cucumber plants.

Monga (1990), tested 12 fungitoxicants against conidial germination and mycelial growth of *A. alternata*, among them Captafol, Copper oxychloride,

Thiram and Mancozeb proved most promising.

Khan and Ahmed (1997), observed the Mancozeb and Propineb decreased fungal growth of *A. alternata* by 76.0 per cent and 72.0 per cent respectively.

Bedi *et al.* (1993), found in laboratory tests and Mancozeb were quite effective against *A. alternata*.

Giri and Presney (1993), evaluated the efficacy of nine fungitoxicants under laboratory conditions and four of these viz., Carbendazin, Fosetyl-Al, Iprodione and Mancozeb inhibited spore germination and mycelial growth of *A. alternata* causing leaf spot disease of Mung bean (*Vigna radiata*, L.).

Paul and Mishra (1993), tested several fungitoxicants *in vitro* for their ability to inhibit the growth of fungi isolated from *Zea mays* seed. Thiram and Captan at 200 ppm. gave maximum inhibition of *A. alternata*. The least effective fungitoxicant in their test was Benomyl.

Shahda *et al.* (1995), found that Etridiazole (Asbansot), Carbendazin and Topsin-M were effective in inhibiting mycelial growth of *A. alternata*.

Vishwakarma and Pandey (1995), reported that among the fungitoxicants tested against *A. alternata* causing Leaf spot of Brinjal *in vitro*. Chlorothalonil, Thiram and Mancozeb completely inhibited the spore germination at 500 ppm. Thiram and Copper oxychloride, were effective in inhibiting the radial growth of the fungus at 1000 ppm. followed by Captan and Captafol.

(ii) SEED TREATMENT

Seed treatment with fungicides is found to very effective in control of seed borne inoculum. Gentner (1923), found reduced infection of *Alternaria* sp. in seed treatment with Gamosan, Mercuran and Thiram. Mills (1975), observed in treated seeds significant increase in germination percentage. Black (1946), was successful to protect seed germination up to 3-5 years by treating the seeds with Ceresan wet or dry stored at 55.0°C-75°C and 50.0 per cent relative humidity.

Dharmvir (1972), found positive results against *Alternaria alternata* of *Triticum vulgare* by seed treatment with Agrosan G.N., Ceresan and Thiram at 0.25 per cent seed weight. Agrosan G.N., Captan, Ceresan and Hexasan eliminated all the fungi including *Alternaria alternata* associated with the seeds and pods of *Arachis hypogea*.

Thiram and Captan, were observed as best seed dressing fungicides to

control the growth of *Alternaria alternata* associated with seeds of Sunflower (Jhamaria *et al.*, 1974). Kumar and Urs (1976), observed Dithane M-45, Difolatan and Thiram in eradication of seed borne mycoflora of *Helianthus annuus*. Narain (1978), found complete control of *Alternaria alternata* in laboratory test by treatment of Sunflower seeds with Agrosan G.N. and Ceresan. Seed treatment with fungicide, Fytolan gave the highest yield (Bhaskaran and Kandaswamy, 1977).

Shukla *et al.* (1980), reported that Agrosan GN, Thiram, Benlate and Dithane M-45 used as seed dressing fungicides on triticales seeds controlled the seed borne infection of *A. alternata* most effectively.

Narain and Saksena (1981), found Agrosan GN, and Ceresan dry effectively eliminated the seed borne infection of *Alternaria alternata* with better seed germination of Sunflower.

Thakur *et al.* (1981), observed significant pre and post-emergence losses in seeds of *Raphanus sativus* infected with *Alternaria alternata*, *Aspergillus niger* and *Cladosporium cladosporioides* and found effective control of seed borne fungi with 0.20 per cent Agrosan G.N. and Ceresan dry.

Vishunavat and Shukla (1982), examined eleven seed dressing fungicides against lentil seed mycoflora and found the elimination of the fungi including *Alternaria* by only Captan. The seeds treated with Dithane M-45 (Mancozeb) or Miltax induced high germination percentage by Prasad (1983). Similarly Jain *et al.* (1983), found in treatment with Bavistin and Captan as best and effective in control of pre and post-emergence seed rot caused by pathogens viz., *Alternaria alternata* and *Curvularia lunata*.

Prasad (1982), found the among the 46 fungi isolated from *Coriandrum sativum*, the most pathogenic one was *A. alternata*. In this study seeds treated with Dithane M-45 (0.05 per cent) or Miltax (0.05 per cent) gave high percentage of seed germination.

Gupta *et al.* (1984), suggested Agrosan G.N., Dithane Z-78 and Dithane M-45 as seed protectants for Pigeonpea and Lentil.

The most promising treatment was achieved by soaking the seeds for 1-2 hours in either Ceresan (Phenyl mercury acetate) or Duter (Penten hydroxide) by Padaganur and Basavaraj (1984). Kumar and Patnaik (1985), observed that seed treatment with Bavistin (Carbendazin) and Difolatan 80-W (Captafol) was

effective in control of pathogenic attack of *Alternaria alternata* and improvement in seed germination by 8.50-24.50 per cent.

Alternaria alternata invading 78.0 per cent seed samples of *Helianthus annuus*, was effectively arrested by Aureofungin, Blitox and Vitavax. Benlate was less effective (Vijaya Lakshmi and Rao, 1985 and Kumar and Patnaik, 1986) during the study of fungicidal treatment of *Alternaria alternata* infected with Pigeon pea, seven fungicides tested significantle. Aureofungin significantly improved seed germination, seedling length, root length shoot length and dry weight.

Mali and Joi (1985), found Captafol, Thiram and Vitavax as most effective as seed dressing fungicide against *Alternaria alternata*.

Shrotri *et al.* (1985), observed that *Alternaria alternata* present in Dahlia seeds was successfully controlled by application of Ceresan.

Kumar and Patnaik (1986), during the study of fungicidal treatment of *Alternaria alternata* infected *Cajanus cajan* only seven fungicides examined significantly improved seed germination, seedling length, root length, shoot length and dry weight.

Prasad and Das (1986), done the seed treatment with 0.50 per cent Ceresan, which inhibited the growth of *Alternaria alternata* and improved the seed germination, plant stand, plump seed percentage and seed yield of *Helianthus annuus*.

Lokesh *et al.* (1987), obtained improved seed germination and seedling vigour of *Cajanus cajan* plants affected by *Alternaria* by seed treatment with Brassicol, Captan and Thiram.

Sharma *et al.* (1987), evaluated the efficacy of twelve fungicides as seed treatment and noticed that Thiram, Emisan and Carbendazim were highly effective against *A. alternata*, *A. padwicki* also improved germination.

Hopper *et al.* (1989), also studied evaluation of several fungicides on seedling emergence and stand establishment of high plain Cotton.

Ram and Maheswari (1992), recorded that *A. alternata* inhibited the germination of Bottle gourd seeds but Difolatan (0.20 per cent) was the most effective one of the five fungitoxicants against *A. alternata* as seed treatment.

Effect of fungicidal seed treatment on seed viability and mycoflora during

storage of *Sorghum* seeds was studied by Solanki (1997).

FIELD EVALUATION OF FUNGICIDES FOR THE CONTROL OF DISEASE -

Spraying of fungicides on plants is found effective to control secondary spread of disease. Presely (1959), recorded Copper oxychloride to be best in control of *Alternaria alternata*, without any phytotoxic effects. Thiram and Zineb were proved to be most effective against *A. alternata* as tested by Misra *et al.* (1970).

Dithiocarbamates viz., Dithane M-45, Dithane Z-78, Cuman, Bisdithane, RH-539, Plantavax and Sulphur dust, were found effective in reduction of incidence of *A. alternata* (Nene *et al.* 1970; Sankhala *et al.* 1972 and Sharifi Tehrani, 1973).

Singh and Singh (1971), reported that Dithane M-45 (0.25 per cent) and Dithane Z-78 (0.25 per cent) were more effective in reduction of disease intensity of *Alternaria* blight of wheat with corresponding increase in yield.

Sharafi Tehrani (1973), recorded that out of the five fungicides examined in the field against *A. alternata* causing Brown spot of Sunflower, Captan and Dithane M-45 (each 0.30 per cent), when sprayed twice at 14 days interval gave the best control of disease.

Agrawal *et al.* (1976), found that application of Dithane M-45 or Dithane Z-78 twice @2 Kg./ha. was most effective against leaf blight of Wheat caused by *Alternaria triticina*. Subsequently spraying of Dithane M-45 (0.20 per cent) and an antibiotic, Aureofungin (0.10 per cent) against *Alternaria* leaf blight of *Helianthus annuus* was observed, which significantly controlled the disease (Sindhamathan *et al.* 1976). *Alternaria* blight of Sunflower was best controlled by Miltox followed by Benlate, Dithane M-45 and Captan (Abraham *et al.* 1976). Reddy and Gupta (1976), examined efficacy of fungicides *in vitro* against *Alternaria helianthi* causing leaf blight of Sunflower.

The effective control of *Alternaria* leaf spot of Mustard was made by Gupta *et al.* (1977) by treatment with Brestan-60, followed by Miltox, Dithane Z-78 and Dithane M-45. Benlate and Aureofungin, were found ineffective (Gupta *et al.* 1977). Kuthubutheen and Pug (1978), observed reduction in growth rate and spore germination of *Alternaria alternata* by treatment with Thiram in comparison to inorganic and mercury fungicides. Narain (1978), found that

among the spray fungicides Captafol and Dithane M-45 followed by Zineb proved to be the best fungicides in eradication of leaf spot of Sunflower caused by *A. alternata*.

Alternaria helianthi was controlled by Bavistin, Dithane M-45, Thiram and Vitavax as most suitable fungicide in *vitro* test in reducing the incidence of disease than Benlate (Bhaskaran and Kandaswamy, 1979 and Mukewar and Gera 1980).

Alternaria helianthi causing Alternaria blight of Sunflower, was effectively controlled by treatment with Mancozeb (0.30 per cent) at interval of 7-10 days enhancing grain yield up to 43.0- 65.0 per cent (Kolte *et al.* 1979), while Bhaskaran and Kandaswamy (1979), found Dithane M-45 and Ziram as highly effective followed by Dithane Z-78, Fytolan and Macubrase.

The comparative efficacy of various fungicides was tested against Alternaria blight of Sunflower by Mukewar and Gera (1980), who find Bavistin, Dithane M-45, Thiram and Vitavax to be the best in reducing the incidence of disease than Benlate, while Saksena *et al.* (1979), found out of the six fungicides Dithane Z-78 (Zineb) and Zineb Ciba, were found to give best results. Agrawal *et al.* (1980). Out of eight fungicides examined, Bavistin and Benlate, followed by Tecto-90, were found most effective to control Alternaria leaf spot of Sunflower. Profic Alwasiak and Szezygiel (1980), employed Captatol, Mancozeb, and Metram four times between flowering and fruit harvest of interval of 8-10 days on Gooseberry and complete eradication of *Alternaria alternata*.

Narain and Saksena (1981), observed Captafol (0.20 per cent) and Dithane M-45 (0.20 per cent) followed by Zineb concluded as best one fungicide for eradication of leaf spot of Sunflower. Rao and Raj Gopalan (1982), was able to control *Alternaria helianthicola* by treatment with fungicides Zn-Mn based on dithio-carbamates on growth and spore germination.

Different fungicides were found to control leaf spot disease of different crops caused by *Alternaria* sp. i.e. Captafol (Rajgopal and Vidyasekaran, 1982) on Tomato caused by *Alternaria solani*, Brestanol (Mathur and Sarbhoy, 1983) on Sugarbeet caused by *Alternaria alternata* and Dithane M-45 (Singh and Shukla, 1984) on Brinjal caused by *Alternaria alternata*.

Tu (1983), found that in the field, Iprodione was the most effective fungitoxicant for controlling both White and Black pod on White beans (*Phaseolus vulgaris*, L.) caused of *A. alternata*.

Tripathi *et al.* (1983), found Captafol followed by Mancozeb and Thiophanate methyl against *Alternaria* leaf spot of Sunflower adducing best effects. Patel *et al.* (1983), observed most effective control of *Alternaria alternata* on Cotton with four sprayings of 0.20 per cent Difolatan (Captafol) at interval of 20 days after the development of disease. Complete control of *Alternaria alternata*, was recorded by Issa (1983) at interval of 15 days in field trials by using four sprayings with Maneb. However Carta *et al.* (1983), found promising control of *Alternaria alternata* by the application of Maneb, Captafol, Chlorothalonil and Iprodione.

Air borne inoculum of *Alternaria alternata*, was controlled by spraying of Dithiocarbamates (Chaudhary *et al.* (1984). Ungaro and Azavedo (1984), recorded Zineb, Priarylphosphite as potential fungicide against *Alternaria alternata* as recorded by Vasu and Rao (1985).

Crisan (1984), observed that Vitavax, Quinolate, Aericide (Binapacry) and Dichlofluanid had a strong fungicidal action against *A. alternata* in Potato, Tomato and beans (*Phaseolus vulgaris*).

Singh and Shukla (1984), reported that *Alternaria* leaf spot and Fruit rot of Brinjal cause of *A. alternata*, was controlled by Dithane M-45 (0.20 per cent) and Brestan -60 (0.10 per cent) at 15 days interval.

Issa (1985), observed the effective control of *A. alternata* causing *Alternaria* disease of Bean by spraying Chlorothalonil, Captafol and Mancozeb alternated with Captafol.

Singh and Shukla (1985), tested several fungitoxicants and found Brestan -60 (0.10 per cent), Captafol (0.20 per cent) and Zineb (0.20 per cent) gave the best control of *Alternaria* leaf spot and Fruit rot of Brinjal caused by *A. alternata* and also increased the yield. The best control of leaf spot of Sunflower was achieved by spray of Dithane M-45 (0.25 per cent) and Carbendazin (0.02 per cent) by Patil and Jadhav (1985).

Singh and Sharma (1986), reported the best results against *A. alternata* causing *Alternaria* fruit rot of Tomato with two sprays of Topsin M and Dithane M-45. Farreta *et al.* (1986), recorded during field trial against Bunch rot of Grapewine caused by *Alternaria* sp., *Cladosporium* sp., *Mucor* sp., *Penicillium* sp. and *Rhizopus* sp., Captafol reduced percentage of damaged cluster by 30.0 per cent and disease severity up to 50.0 per cent. In an experiment conducted for

three years Blitox (Copper oxychloride), Cuman L. (Ziram), Dithane M-45 (Mancozeb) and Duter reduced the disease index of leaf spot caused by *Alternaria macrospora* in Upland Cotton, C.V. Laxmi and C.V. M.C.U-S. Cuman (0.40 per cent) and Blitox (0.30 per cent) gave the best control (Padaganur and Basavaraj, 1987).

Al-Samdisy *et al.* (1987), found that Dithane M-45 (0.20 per cent) was the most effective fungitoxicant against *A. alternata* affecting Faba bean.

Thind and Jhooty (1987), reported that among 12 fungitoxicants tested the most effective and economical control of *A. alternata* causing Black rot of Chillies under Punjab conditions, was given by sprays of Captafol (0.20 per cent) followed by Dithane M-45 (0.20 per cent), Cuman-L (0.30 per cent), Blitox (0.30 per cent) and Captan (0.20 per cent).

Cpatafol and Dithane M-45 proved to be effective fungicide in control of Purple blotch of Onion caused by *A. porri* and blight of Fennel caused by *A. alternata* and increased the yield significantly (Sharma, 1986 and Chaudhari and Patel, 1987).

Kumar and Pandey (1988), reported the best control of leaf of Onion caused by *A. alternata* in U.P. was given by four applications of Dithane M-45 (0.25 per cent) at 20 days interval with 65.50 per cent disease reduction followed by Carbendazim (0.10 per cent with 59.10 per cent).

Kakade and Khune (1989), studied the efficacy of fungicides and found 2-Thiocyanomethylthio benzothiozole to be most effective. Calixin inhibited also the growth of *Alternaria alternata*.

Ample control of *Alternaria* leaf spot of Sesame caused by *A. alternata* was achieved by three sprayings of Mancozeb (0.25 per cent), Captan (0.20 per cent) and Carbendazim (0.10 per cent) at 15 days interval.

Mancozeb (0.2 per cent) was found best fungicide in controlling *Alternaria* blight of Onion by (Vishwakarma, 1990).

Raut and Sman (1990), found that in field trials during 1982- 1985 the incidence of Pod blight of Gram caused by *A. alternata* was reduced by spray of Chlorothanolil, Dithane M-45 (Blitox-50), Ziram and Rovral (Iprodione) with maximum disease reduction occurring when Carbendazim or Callixin were applied starting from first incidence of the disease alongwith the highest

increase in seed yield. Mancozeb (0.2 per cent) was found as best fungicide for the control of *Alternaria* blight of Onion (Vishwakarma, 1990).

Reddy *et al.* (1991), studied fungicidal control of *Alternaria* blight of Sunflower. Bhardwaj (1991), reported that sequential application of Captafol (0.25 per cent), Mancozeb (0.25 per cent) and Copper oxychloride (0.25 per cent) in 40, 55 and 70 days after transplanting, increased yield by 50.50 per cent alongwith reduction in incidence of *A. solani* and *A. alternata* in Tomato. The net gain per rupee invested was Rs. 21.52. The spraying with Captafol (0.25 per cent) alone gave the highest net income but the benefit per rupee was only Rs. 16.13

Castro *et al.* (1991), tested the efficacy of eleven fungitoxicants of two cultivars of *Phaseolus vulgaris* eg. Carioca and Carioca, 80, naturally infected by *Alternaria* sp. Among these Chlarothanonil, Mancozeb, Tri-phenyltin acetate gave satisfactory control of this disease).

Maheswari *et al.* (1991), tested six compounds in field trial during 1889-1891 and recorded the most effective control (64.70 per cent) of *A. solani* causing Early blight of Tomato was given by Copper oxychloride followed by Mancozeb (61.70 per cent). The highest increase in yield was recorded in plots sprayed with Mancozeb.

Monga (1991), made observation on the chemical control of Brown spot of Tobacco caused by *A. alternata*. The disease was controlled by two sprays of Thiram (0.2 per cent) or Mancozeb (0.2 per cent) once after appearance of the disease and again 10 days later.

Barros *et al.* (1992), carried out study to determine the effect of number of sprays of Mancozeb (2 Kg. per hectare) at different periods for control of *Alternaria* leaf spot of Bean (*Alternaria* sp.) cultivars Carioca and Carioca-80 during 1987-1988. The best yield and control, were obtained when seven sprays with Mancozeb, were applied beginning from 20 days after plant emergence at 10 days interval. Satisfactory disease control was achieved by spraying the plants 20, 35 and 50 days after the plant emergence.

Khan and Ahmad (1992), tested Propineb (Antracol), Benomyl, Copper oxychloride and Mancozeb for controlling *Alternaria* leaf spot of Potato, Copper oxychloride and Mancozeb decreased intensity up to 72.0 and 60.0 per cent and increased Potato yield by 80 and 55 per cent compared with untreated crops.

Tu (1992), screened several fungitoxicants as spray but only Iprodione (10.2 per cent) was the most effective for controlling Alternaria black pod disease of Bean caused by *A. alternata* and foliar spraying was recommended just prior to plant senescence.

Efficacy of fungicides in initiation of chemical control of Alternaria leaf spot in Pima Cotton, was studied by Shtienberg (1992). Bio-efficacy of insecticides in combination with fungicides against Alternaria leaf spot of Cotton was made by Underwade *et al.* (1993).

Lonsdale and Kotze (1993), reported that in field trials, blossom sprays with Iprodione, Chinomethionate, Triadimenol, Copper oxychloride, Flusilazol, Mancozeb and Pysazophos, were evaluated over two seasons during 1989-91 for the control of Blossom spot disease of Mango caused by *A. alternata*. Among these Flusilazol was the most effective chemical against this disease. A mixture of Flusilazol + Mancozeb gave better control than flusilazol alone.

Kumar and Singh (1994), found the best results in control of Leaf spot of Sunflower by treatment with Dithane M-45 and Breston-60 at the rate of 20.0 per cent at an interval of 10 days.

Foliar application of Fungicide and Potassium in the late season on leaf spot caused by *Alternaria sp.* was examined by Newman and Howard (1995).

Vishwakarma and Pandey (1995), found that Mancozeb (0.2 per cent) sprayed four times at 10 days intervals, was superior among all the fungitoxicants tested, which reduced the severity of leaf spot of Brinjal caused by *A. alternata* by 65.26 per cent over control. The next best fungicides were Dithianom Thiram, Folpet and Zineb, where disease severity varied from 63.09 to 63.91 per cent. Significant increase in yield (129.0 to 197.29 per cent), was achieved in field sprayed with Mancozeb, Zineb, Dithianom, Thiram and Chlorothalonil.

Narain (1995), observed that Dithane M-45 (0.20 per cent) was the most effective fungicide in controlling the Alternariosis caused by *A. alternata*, *A. cyamopsidis*, *A. zinniae* and *A. tenuissima* in five beans namely broad bean, cluster bean, french bean, soybean and winged bean and four vegetables eg. Bathua, Lettuce, Methi and Spinach.

Singh *et al.* (1995), reported that amongst four fungicides viz., Indofil M-45 (0.25 per cent), Rovral (0.25 per cent), Calixin (0.10 per cent) and Suflex (0.25 per cent), Rovral was found to be most effective for controlling Alternaria blight of linseed followed by Indofil M-45. The maximum disease control and the highest yield was recorded with three sprays of Rovral. Management of Alternaria leaf spot of cotton in detail was studied by Khan *et al.* (1999).



Chapter - III

MATERIAL AND METHOD

MATERIAL AND METHOD

1. SURVEY AND COLLECTION OF DISEASED MATERIALS FROM THE DIFFERENT LOCALITIES :-

To find out the prevalence and severity of Dolichos bean (*Dolichos lablab*, L.) leaves infected with leaf spot caused by *Alternaria alternata* (Fries.), Keissler, an extensive survey was made during the seasons of 2001 and 2002 from Vegetable Research Farm Kalyanpur, Kanpur, G.S. Pant University Farm Pant Nagar and different research stations of Uttar Pradesh, where the crop is sown as given in Table - 1.

Table - I

Particulars of different Research Stations of U.P. regarding prevalence and severity of leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler.

S.No.	Research Station	Location
1.	Agriculture Science Centre	Ganiwa Chirakoot
2.	Charyar Farm	Bulandshahar
3.	Crop Research Farm	Etawah
4.	Regional Research Station	Saini, Allahabad
5.	Crop Research Farm	Saraimira, Farrukhabad
6.	Crop Research Farm	Baraur, Farrukhabad
7.	Crop Research Farm	Araul, Kanpur
8.	Crop Research Farm	Deegh, Kanpur
9.	Crop Research Farm	Modipuram Area, Meerut
10.	Crop Research Centre	Mauranipur, Jhansi
11.	C.S. Azad University Research Farm	Belatal, Mahoba
12.	Directorate of Vegetable Research Farm	Varanasi

13.	G.P. Pant University Farm	Pant Nagar, Nainital
14.	Govt. Agriculture Centre	Atarra, Banda
15.	Government Agriculture Centre	Bansi, Banda
16.	Government Agriculture Centre	Khaptiha, Banda
17.	Government Agriculture Research Centre	Bharari, Jhansi
18.	Government Agriculture Centre	Bagi, Jalaun
19.	Groundnut Research Station	Mainpuri
20.	N.D. University	Kumarganj, Faizabad.
21.	Oil Seed Research Farm	Kalyanpur, Kanpur
22.	Regional Research Centre	Amroha, Jhansi
23.	Regional Research Station	Uttaripura, Kanpur Dehat
24.	Regional Research Centre	Hardoi
25.	Regional Research Centre	Varanasi
26.	Regional Research Station	Dilip Nagar, Kanpur
27.	Regional Research Station	Madhurikund, Mathura
28.	Regional Agricultural Testing Demonstration Centre	Meerut
29.	Vegetable Research Farm	C.S. Azad University of Agriculture and Technology, Kanpur.
30.	Vegetable Research Farm	Kalyanpur, Kanpur

Naturally infected leaves of Dolichos bean showing the characteristic symptoms of the disease leaf spot were collected, brought in the laboratory and critically examined macroscopically and microscopically for the study of symptomology and causal organism. The specimens used for isolation of pathogen in culture, were preserved, labelled and kept in dry and wet forms for further investigations and records.

2. METHOD FOR RECORDING DISEASE INCIDENCE AND DISEASE INTENSITY.

(A) DISEASE INCIDENCE -

The incidence of disease was recorded in the field by randomly locating five sub-plots of 10.0 square meters and counting the number of diseased and healthy plants of Dolichos bean. The average disease incidence was calculated in percentage by the method suggested by Chester (1950), as given below.

$$\text{Disease incidence} = \frac{\text{Number of diseased plants affected in total population}}{\text{(samples collected)}}$$

$$\text{Percent disease incidence} = \frac{\text{Total number of diseased plants}}{\text{Total number of plants in the field}} \times 100$$

(B) DISEASE INTENSITY -

One hundred affected leaves were randomly selected from each field, which were arranged in six grades from 0-5 on the basis of percent lamina area affected according to the method given by James (1971) and Chopra and Sharma (1976). The disease categories were established as given in Table II.

Table - II

Disease categories on the basis of leaf area affected.

Percent leaf area (affected)	Grade	Number of leaves in grade (in ratings)	Disease ratings
0	0	N ₀	N ₁ × 0
5	1	N ₁	N ₂ × 1
10	2	N ₂	N ₃ × 2
20	3	N ₃	N ₄ × 3
30	4	N ₄	N ₅ × 4
40 and above	5	N ₅	N ₆ × 5

Numerical Ratings

0 = No infection.

1 = Up to 5.0 per cent leaf area affected.

2 = Up to 10.0 per cent leaf area affected.

3 = Up to 20.0 per cent leaf area affected.

4 = Up to 30.0 per cent leaf area affected.

5 = Up to 40.0 per cent leaf area affected.

The per cent disease index was calculated according to the formula given below :-

$$\text{Disease intensity} = \frac{N_1 \times 0 + N_2 \times 1 + N_3 \times 2 + N_4 \times 3 + N_5 \times 4 + N_6 \times 5}{N \times 5} \times 100$$

N = Total number of leaves examined.

N₀ to N₆ = Number of leaves in different grades.

3. SYMPTOMS OF THE DISEASE UNDER NATURAL CONDITIONS :-

The disease symptoms on the leaves observed under natural conditions of infection, were carefully studied and shape, size, colour and development of spots were recorded.

4. ISOLATION AND PURIFICATION OF PATHOGEN -

The leaf spots depicting initial and distinct characteristic symptoms on the leaves of different affected plants of Dolichos bean, were collected in fresh polythene bags for the purpose of isolation of pathogen in pure culture. The selected leaves alongwith infection were plucked and after the macroscopic and microscopic examination, were thoroughly washed with sterilized water in order to remove dust particles and surface contaminants. Subsequently infected host tissues or young diseased part of leaves were cut into small bits of about 2.0 mm. long alongwith some healthy tissues with the help of sterilized scalpel.

These leaf bits were washed in 3-4 changes of sterilized water in order to remove the disinfectants and thereafter dipped in 0.10 per cent solution of mercuric chloride for 30 seconds with the help of sterilized forceps and washed thoroughly with 3-4 changes of sterilized water to remove the traces of mercuric chloride. Excess moisture was removed by placing these pieces in between two folds of sterilized blotting papers under aseptic conditions in the inoculation chamber. These surface sterilized pieces, were transferred with the help of sterilized forcep to petridish poured with 2.0 per cent Potato dextrose agar medium. In each petridish leaf bits were placed at equal distances and incubated for 10 days at 25±1°C.

The petridishes used for the purpose of isolation, were sterilized in hot air oven at 160°C for two hours and Potato dextrose agar (P.D.A.) medium was also sterilized in autoclave as 15 lbs. pressure (p.s.i.) for twenty minutes.

After 24 hours of incubation as soon as the mycelial growth became visible around these pieces the hyphal tips from advancing mycelium, were transferred aseptically to the sterilized culture tubes containing Potato dextrose agar (P.D.A.) medium and further purified by single spore culture technique suggested by Keitt (1915). For this estimation dilution method was followed and counts were made under microscopic field under low power (10 X). For this purpose one randomly flask having fungal growth was thoroughly shaken for each medium. From these flasks one ml. fungal spore suspension was taken and diluted five times with distilled water. One ml. of this diluted solution was further divided into five equal drops on glass slides for conidia of fungus under low power (10 X) of microscope. Their numbers were then counted in each drop. Finally the average number of conidia was calculated by dividing the total number of five and categorization was made on the basis of scale given in Table III as below :-

Table - III

S.No.	Number of spores per microscopic field	Symbol	Intensity of Sporulation
1.	Nil	—	Absent (Nil)
2.	1 – 5	+	Poor
3.	6 – 10	+	Fair
4.	11 – 15	+++	Good
5.	16 and above	++++	Excellent

A dilute spore suspension was poured in plain agar petridishes to settle down on the agar medium. The amount of spore suspension, was adjusted so that a very thin layer could be formed over surface of agar medium. The spores which settled quite apart from the other, were observed under the microscope, which were marked and encircled on the back of bottom of petridish with a glass marking pencil, were lifted alongwith agar medium with the help of dummy cutter in petridish and transferred to petridishes containing Potato dextrose agar (P.D.A.) medium. Regular sub-culturing was done to check further growth of contaminants from growth obtained with single spore culture technique.

The pure culture of the pathogen thus obtained was multiplied and maintained at 2.0 per cent Potato dextrose agar (P.D.A.) medium slants in the B.O.D. Incubator at 8°C - 10°C for further cultural and morphological studies and pathogenic behaviour of pathogen. The cultures were multiplied and revived after every two months.

5. PATHOGENICITY TEST :-

The pathogenicity of the test fungus isolated from the affected leaves was made on the healthy leaves of host plants, in order to establish the pathogenic nature of fungus according to Koch's Postulates. For this study the plants were raised in earthen, pots of 30.0 cm. diameter filled with sterilized soil by sowing seeds of variety "Kalyanpur Type-1", which was recorded to be highly susceptible to leaf spot disease under natural conditions and five plants per pot were maintained. Few leaves were inoculated by spraying spore-cum-mycelial suspension prepared in sterilized water with the help of an atomizer. Two hundred leaves under each treatment were mechanically injured and uninjured were employed for inoculation purposes. After inoculation the potted plants were kept in moist chamber for 48 hours to provide optimum humidity for infection by covering them under polythene bags. A separate set of plants was left un-inoculated to serve as control by spraying only sterilized water in place of fungal suspension.

After inoculation all the potted plants tested including control, were shifted to glass house benches, where they were watered periodically to maintain sufficient humidity for proper growth of plants and development of disease. These plants were examined periodically to record the symptoms and final data on disease were recorded after fifteen days of inoculation. The fungus was reisolated from the artificially inoculated leaves and compared to original isolate of pathogen. The experiment was replicated five times.

6. IDENTIFICATION OF THE PATHOGEN :

The pathogen, *Alternaria alternata* (Fries.), Keissler, was identified on the basis of morphological and cultural characters as well as pathogenic behaviour of host. The morphological and cultural characters of the isolated fungus was studied on Potato dextrose agar (P.D.A.) medium which exhibited the best growth of fungus. For identification, Ridgway (1912) "Colour Standard and Colour Nomenclature", was used for determining the colour production by the pathogen in different types of studies.

Poured petridishes, were inoculated with the isolated fungus under study and the cultural characters viz., type and colour of growth on Potato Dextrose agar (P.D.A.) medium and morphological characters viz., colour; width and branching of mycelium; shape; size; septation; septation of beak, chain formation and colour of conidia as well as chlamydospores were recorded after incubation for ten days at room temperature $25\pm 1^{\circ}\text{C}$.

(A) MYCELIAL AND COLONY CHARACTERS -

On 2.0 per cent Potato dextrose agar (P.D.A.) medium was used for the study of colony and mycelial characters of the pathogen. Four petridishes were inoculated with the fungal isolate the *Alternaria alternata* (Fries.), Keissler and following characters were examined -

(a) COLONY CHARACTERS

1. Colour
2. Type of Growth

(b) MYCELIAL CHARACTERS

1. Colour of hyphae
2. Branching of hyphae
3. Septation of hyphae
4. Width of hyphae

(B) CONIDIOPHORE CHARACTERS

1. Colour
2. Shape
3. Size
4. Septation

(C) CONIDIAL CHARACTERS

1. Colour
2. Arrangement
3. Shape

4. Size
5. Septation
6. Surface

(D) BEAK

1. Length
2. Width
3. Septation

(E) CHLAMYDOSPORES

1. Shape
2. Colour
3. Size

Slides for the study of fungal characters including hyphae, conidia and conidiophores, were prepared from 10 days old cultures selected at random. Temporary slides were prepared in Cotton blue.

7. CULTURAL STUDIES OF THE PATHOGEN

Cultural studies of pathogen, was made on the different natural (non synthetic, semi-synthetic and synthetic media to investigate the growth and sporulation of the pathogen isolated and thereafter the best suited medium for fungal growth and sporulation, which are as follows -

(A) NATURAL (NON-SYNTHETIC) MEDIA :-

1. Potato Dextrose Medium

Pealed and Sliced potatoes	200.0 gms.
Agar Agar	20.0 gms.
Dextrose	20.0 gms.
Distilled water	1000.0 ml.

2. Corn Meal Medium

Corn Meal	50.0 gms.
Distilled water	1000.0 ml.

3. Leaf Decoction Medium

Chopped Dolichos bean leaves	200.0 gms.
Distilled water	1000.0 ml.

(B) SEMI-SYNTHETIC MEDIA :-

1. Malt Salt Medium

Malt Extract	20.0 gms.
Salt (NaCl)	10.0 gms.
Distilled water	1000.0 ml.

2. Sabouraud's Medium

Glucose	40.0 gms.
Peptone	10.0 gms.
Distilled water	1000.0 ml.

(C) SYNTHETIC MEDIA :-

1. Coon's Medium

Potassium nitrate	2.0 gms.
Magnesium sulphate	1.20 gms.
Potassium dihydrogen phosphate	2.70 gms.
Maltose	7.20 gms.
Distilled water	1000.0 ml.

2. Czapek's (Dox) Medium

Magnesium sulphate	0.50 gms.
Potassium dihydrogen phosphate	1.0 gms.
Potassium chloride	0.50 gms.
Ferrous sulphate	0.01 gms.
Sodium nitrate	2.0 gms.
Sucrose	30.0 gms.

Distilled water	1000.0 ml.
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3. **Richard's Medium**

Potassium nitrate	10.0 gms.
Potassium dihydrogen phosphate	5.0 gms.
Magnesium sulphate	2.50 gms.
Ferric chloride	0.02 gms.
Sucrose	50.0 gms.
Distilled water	1000.0 ml.

4. **Asthana and Hawker's Medium**

Potassium nitrate	3.50 gms.
Magnesium sulphate	0.75 gms.
Potassium hydrogen phosphate	1.75 gms.
Glucose	5.0 gms.
Distilled water	1000.0 ml.

5. **Brown's Medium**

Tripotassium phosphate	1.25 gms.
Magnesium sulphate	0.75 gms.
Asparagine	2.0 gms.
Glucose	20.0 gms.
Distilled water	1000.0 ml.

For use of medium in solid form 20.0 gms., Agar Agar was added apart from the above ingredients in each medium.

(a) **PREPARATION OF MEDIA :-**

The different kinds of media used in experiments in the present investigation, were prepared by the standard methods as described by Riker and Riker (1936). The various ingredients of media, were dissolved in distilled water separately before mixing together and for solidification 2.0 per cent Agar Agar

was mixed, wherever necessary. In case of Potato Dextrose, Corn meal, Oat meal and Host leaf, extracts were taken after boiling them on water bath. All the media were sterilized in Autoclave 15.0 lb. pressure (p.s.i.) for 20 minutes.

(b) CLEANING AND STERILIZATION OF GLASSWARES :-

All the glasswares such as petridishes, pipettes, funnels, flasks and glass rods, were cleaned in Chromic acid (Potassium dichromate) solution 10.0 gms, Sulphuric acid 60 ml. and water 1000.0 ml. followed by thorough rinsing in flowing water. After drying the petridishes were sterilized at 160°C for one year in an electric oven.

(c) FUNGAL INOCULUM :-

Ten days old culture of the pathogen under investigation grown on 2.0 per cent Potato dextrose agar (P.D.A.) medium in sterilized petridishes, was used as inoculum for all the studies. Petridishes containing equal amount of P.D.A. medium were inoculated with 5.0 mm. culture discs cut with the help of sterilized cork borer from the growing region of the pathogen. The flasks containing liquid media, were also inoculated aseptically with 5.0 mm. culture discs of the pathogen.

(d) SELECTION OF BASAL MEDIUM :-

For the selection of basal medium the pathogen was grown on different solid and liquid media belonging to natural (non- synthetic), semi-synthetic and synthetic types. The average mycelial radial growth, mycelial dry weight and sporulation of the pathogen, were considered as main criterion for the selection of basal medium. Five sets of petridishes and flasks were used respectively in all the experiments.

8. DETERMINATION OF FUNGAL GROWTH :-

(i) RADIAL GROWTH

To study the radial growth of the pathogen 20.0 ml. of sterilized medium was poured into sterilized petridishes and after the solidification of medium equal discs of 5.0 mm. diameter of the fungal growth cut by the help of cork borer, were placed at the centre of each petridish. After inoculation the petridishes were incubated for 10 days at 25±1°C. Each experiment was replicated five times. After the expiry of incubation period radial growth of pathogen was observed in mm. in two directions at right angles to each other

and thereafter average was calculated. Cultural characters viz., colony type, colour of colony, substrate colour and amount of aerial mycelium were recorded. The colonies of the pathogen observed were categorised into groups as given in table IV.

TABLE IV

Particulars of Measurement of radial growth of pathogen *Alternaria alternata* (Fries.), Keissler

S.No.	Diameter of Colony (mm.)	Type of Growth	Symbol
1.	20 and less	Very poor	—
2.	21 to 40	Poor	+
3.	41 to 60	Fair	+
4.	61 to 80	Good	+++
5.	81 to above	Excellent	++++

(ii) QUANTITATIVE GROWTH :-

For determination of fungal dry weight the fungus was grown in 50.0 ml. liquid media into 150 ml. Erlenmeyers flasks. The flasks were sealed with cotton plugs tightly. These flasks were then sterilized in autoclave at 15 lb. pressure (p.s.i.) for 20 minutes. The flasks were inoculated by equal sized 5.0 mm. discs obtained from the old culture and were incubated at room temperature $25 \pm 1^\circ\text{C}$ for ten days. After the expiry of incubation period the fungal mats obtained from each of the flasks were filtered through whatman's filter paper No. 42 separately. These filter papers alongwith the fungal mats were dried in electric oven at 60°C for 48 hours. Completely dried samples were kept in dessicators over anhydrous Calcium chloride and weighed separately and accurately on analytical balance. The actual dry weight of the fungal mat, was determined by deducting the weight of filter paper from the total weight and of which average was calculated. Five replications were taken as standard value for comparing the growth under different treatments.

(iii) DETERMINATION OF SPORULATION :-

The degree of sporulation was graded in the five categories on the basis of spore count under low power of microscope. For this one flask from each medium was randomly taken and thoroughly shaken. From these flasks 1.0 ml. of fungal suspension was taken and diluted to five times with distilled water. 1.0 ml. of

this diluted suspension was divided into five equal drops and placed in Haemocytometer for counting the spores under low power of microscope. Thus spore counts were taken in each drop of diluted medium having fungal growth. Finally the average number of the spores was calculated by dividing the total number by five and categorisation was made on the basis of scale given in table IV.

9. PHYSIOLOGICAL STUDIES

The physiological investigations including effect of temperature and Hydrogen ion concentration on the growth of pathogen and nutritional requirements, were made by the following standard techniques which are described below.

(a) TEMPERATURE

To study the effect of temperature on growth and sporulation, the pathogen was grown at different temperatures viz., 5°C (T₁), 10°C (T₂), 15°C (T₃), 20°C (T₄), 25°C (T₅), 30°C (T₆), 35°C (T₇), 40°C (T₈), 45°C (T₉), 50°C (T₁₀) in different incubators. In this study Potato dextrose agar (P.D.A.) medium was used as basal medium and methods of sterilization, inoculation, filtration and determination of the dry weight of mycelial weight, were followed as described earlier. To avoid log effects the flasks containing the same medium were incubated at the temperatures above for 24 hours before inoculation. Subsequently inoculation was done and observations on the average, dry mycelial weight and degree of sporulation was determined after 10 days of incubation. Each treatment was replicated three times.

(b) HYDROGEN - ION CONCENTRATION

To study the effect of Hydrogen ion concentration (pH) the Pathogen was grown in 150.0 ml. flasks containing basal medium (Potato dextrose agar medium) as suggested by Clark (1928) on 20 different pH values viz. 2.50 (P₁), 3.0 (P₂), 3.50 (P₃), 4.0 (P₄), 4.50 (P₅), 5.0 (P₆), 5.50 (P₇), 6.0 (P₈), 6.50 (P₉), 7.0 (P₁₀), 7.50 (P₁₁), 8.0 (P₁₂), 8.50 (P₁₃), 9.0 (P₁₄), 9.50 (P₁₅), 10.0 (P₁₆), 10.50 (P₁₇), 11.0 (P₁₈), 11.50 (P₁₉) and 12.0 (P₂₀). Before sterilization initial pH values were adjusted by using N/10 Sodium hydroxide and N/10 Hydrochloric acid and accurately measured by Backman's pH paper. The treatment was replicated three times. After measurement of pH the medium was sterilized in autoclave at 15 lb. pressure (p.s.i.) for 20 minutes as described earlier and again pH values of

each treatment were measured. The pH was adjusted to original values by adding measured quantities of N/10 NaOH or N/10 HCl under aseptic conditions in the culture chamber. After adjusting the required pH values the discs of 5.0 mm. of pathogen were inoculated in each flask, which were incubated at $25\pm 1^{\circ}\text{C}$ up to the time till the growth in any one of flasks was completed. The average dry weight of the mycelium and extent of sporulation was recorded.

10. ENZYMIC STUDIES

In the present investigation a study of comparison in the production of Cellulase (Cx), Polymethylgalacturonase (PMG) and Polygalacturonase (PG) enzymes by pathogen in affected Dolichos bean leaves causing leaf spot as well as in cultures, was made out to find out variations in their production.

(A) PRODUCTION OF EXTRA CELLULAR ENZYMES IN VITRO -

(a) PREPARATION OF ENZYME SAMPLE -

For the study of production of pectolytic and cellulolytic enzymes in *vitro* the pathogen, *Alternaria alternata* (Fries.), Keissler was grown in 150.0 ml. Erlenmeyer's flasks containing 50.0 ml. sterilized Richard's liquid medium by transferring 0.50 mm. diameter mycelial mat discs from 10 days old culture to each flask.

The medium was adjusted at pH 6.0 before sterilization. After inoculation the flasks were incubated at $25\pm 1^{\circ}\text{C}$ for 15 days. The inoculated flasks were vigorously shaken in order to make a uniform suspension after which the fungal growth was filtered through Whatman's filter paper No. 42 twice and the filtrate was allowed to collect. The filtrate was finally centrifuged at 4000 rpm for 20 minutes and clear supernatant was collected in sterilized air tight bottles, while the sediment was rejected. Two ml. of toluene was then added to cover the upper surface of enzyme preparations to provide aseptic conditions. These bottles were then stored at 5°C in a B.O.D. Incubator for use of them as crude enzyme sample for enzyme assay. The enzyme assay was carried out at $25\pm 1^{\circ}\text{C}$ in water bath.

(b) ENZYME ASSAY :

The crude enzyme preparations thus obtained were assayed for the production and activity of the hydrolytic enzymes of both the pectolytic and cellulolytic nature at $25\pm 1^{\circ}\text{C}$ on a water bath.

(i) CELLULASE (CX) ASSAY :-

Cellulase (CX) activity was determined by Viscometric method by measuring loss in viscosity for Carboxymethyl cellulose (CMC) solution, followed by Muse *et al.* (1972). 7.0 ml. of 0.6 per cent Carboxy methyl cellulose (CMC) buffered at pH 5.60, was used as substrate for estimation of enzyme action. Two ml. of crude enzyme sample (culture filtrate) was added to 7.0 ml. of substrate in 0.05 M. aqueous Sodium citrate solution at 7.0 pH placed in Ostwald Fenske Viscometer at 30°C in a water bath according to the technique suggested by Hamcock *et al.* (1964). 10 ml. of enzyme solution was added to this and mixed jointly by drawing air rapidly through the large arm of Viscometer by suction. The time of efflux was recorded immediately at zero hour. Subsequently the readings of efflux time, were recorded for incubation period for 0, 30, 60, 90, 120 and 150 minutes in each separately at 25±1°C. Controls were used with distilled water for comparison. The pH level was adjusted with Mellavain's Citrate Buffer (MC Clavain, 1921).

The reduction in Viscosity, was expressed as percentage loss in viscosity over water (control) as calculated by the formula as given below :-

$$\text{Percent loss in Viscosity} = \frac{T_0 - T_1}{T_0 - T_w} \times 100$$

over control

where,

T_0 = Flow time of reaction mixture at zero hour.

T_1 = Flow time of reaction mixture at a particular interval.

T_w = Flow time of distilled water.

(ii) POLYGALACTURONASE (PG) ASSAY :-

It was determined by same technique as described in cellulase assay except that 3.0 ml. of Sodium polypectate was used in the substrate in place of CMC buffered at pH 4.60

(iii) POLYMETHYLGALACTURONASE (P.M.G.) ASSAY :-

It was determined by the same technique as described in cellulase assay

except that 1.20 per cent Citrus pectin was used in the substrate in place of C.M.C. and buffered at pH 6.0

(B) PRODUCTION OF ENZYMES IN VIVO

The assay of enzymes *in vivo*, was determined by extracting crude enzymes from the leaves of Kalyanpur, variety of Type - 1, Dolichos bean, which were affected by the disease. For comparison healthy leaves of the host variety were used as control. For each sample 7.0 gms. diseased material was grinded in 30 ml. of 0.25 M. Sodium chloride solution. The extract thus obtained was filtered and filtrate was collected and finally centrifuged at 4000 r.p.m. for 20 minutes and the clear supernatant solution collected in sterilized air tight bottles, were used as crude enzyme for further studies. The enzyme assayed were cellulase (CX), Polymethylgalacturonase (PMG) and Polygalacturonase (PG). The technique adopted was the same as followed in *vitro* studies.

II. NUTRITIONAL STUDIES :-

To study the effect of various nutritional sources such as carbon, nitrogen and N.P.K. on the growth and sporulation of the pathogen, the substitutions of these ingredients, were made in basal Potato dextrose agar medium. Before sterilization the pH of the medium in each flask was adjusted to 6.50. The flasks inoculated with an equal quantity of the inoculum were incubated at $25 \pm 1^\circ\text{C}$ for ten days. After the incubation period the average dry weight of the mycelium and the extent of sporulation were recorded. Each treatment was replicated three times.

(a) EFFECT OF DIFFERENT SOURCES OF CARBON ON THE GROWTH AND SPORULATION OF THE PATHOGEN :-

To study the effect of various sources of Carbon on growth and sporulation of the pathogen, these compounds were substituted singly in the carbon-free basal medium bringing the carbon level at par with that contained in 500.0 gms. of sucrose. The medium containing each carbon source separately was dispensed in three 150 ml. flasks in 50 ml. quantities. A carbon free medium served as control.

The methods of sterilization, filtration, determination of sporulation and dry mycelial weight, were followed as detailed earlier. The flasks were incubated

at $25 \pm 1^\circ\text{C}$.

The following carbon compounds were used in this study :-

(A) Monosaccharides

(a) Pentoses

1. D-xylose
2. Rhamnose

(b) Hexoses

1. D-glucose
2. D-galactose
3. D-mannose
4. D-fructose

(B) Disaccharides

1. Lactose
2. Maltose
3. Sucrose

(C) Trisaccharides

1. D-raffinose

(D) Alcohols

1. D-mannitol
2. Sorbitol

(E) Polysaccharides

1. Dextrin

(b) EFFECT OF DIFFERENT SOURCES OF NITROGEN ON GROWTH AND SPORULATION OF THE PATHOGEN -

With a view to study the effects of various nitrogen compounds over the growth and sporulation of the pathogen, these compounds were substituted singly for potassium nitrate in the basal medium. The concentration was so

adjusted as to get the amount of nitrogen present in 10.0 gm. of sodium nitrate per litre of the basal medium. Peptone was however substituted in an amount equal to that of Sodium nitrate. Medium without nitrogen served as control. 50.0 ml. of the medium containing each nitrogen source separately was poured in three 150 ml. flasks. The method of sterilization of medium, inoculation, filtration, determination of dry mycelial weight and sporulation were the same as described for carbon sources. The inoculated flasks were incubated at $25\pm 1^{\circ}\text{C}$ for 10 days before determining the dry mycelial weight and sporulation. The following twelve inorganic nitrogen compounds were used in the investigation.

(A) Organic compounds

1. Peptone
2. Urea
3. Thio-urea

(B) Inorganic compounds -

4. Ammonium chloride
5. Ammonium carbonate
6. Ammonium sulphate
7. Ammonium nitrate
8. Ammonium oxalate
9. Ammonium acetate
10. Calcium nitrate
11. Potassium nitrate
12. Sodium nitrate

(C) EFFECT OF DIFFERENT DOSES OF NITROGEN, PHOSPHORUS AND POTASH ON THE SEVERITY OF DISEASE :-

To determine the effect of different doses of nitrogen, phosphorus and potash on the severity of Alternaria leaf spot of Dolichos bean, an experiment was laid out at the glass house compound of the department during Rabi and Kharif seasons of 2001 and 2002 in randomized block design with three replications in the plot size of 2.0 m. \times 5.0 m. A highly susceptible variety,

"Kalyanpur Type-1" of *Dolichos* bean was sown for this study.

The fertilizer doses with different formulations and combinations included in this experiment are as follows -

Nitrogen level	Phosphorus level	Potassium level
N ₀ - 0 Kg./ha.	P ₀ - 0 Kg./ha.	K ₀ - 0 Kg./ha.
N ₁ - 60 Kg./ha.	P ₁ - 60 Kg./ha.	K ₁ - 40 Kg./ha.
N ₂ - 120 Kg./ha.		

Treatment combinations

N ₀ P ₀ K ₀	N ₁ P ₀ K ₀	N ₂ P ₀ K ₀
N ₀ P ₀ K ₁	P ₁ P ₀ K ₁	N ₂ P ₀ K ₁
N ₀ P ₁ K ₀	N ₁ P ₁ K ₀	N ₂ P ₁ K ₀
N ₀ P ₁ K ₁	N ₁ P ₁ K ₁	N ₂ P ₁ K ₁

The levels of nitrogen, phosphorus and potash, were applied in the form of urea (46.0 per cent nitrogen), single super phosphate (16.0 per cent phosphorus) and muriate of potash (60.0 per cent potash). Half quantity of nitrogen and full quantities of phosphorus and potash, were applied at the time of sowing as the basal dose and the remaining half quantity of nitrogen was applied at top dressing after one month of sowing. No fertilizer was given in control. The disease intensity was recorded on the basis of per cent leaf area affected at harvesting time of the crop.

12. SUSCEPTIBLE GROWTH PERIOD OF HOST -

In order to study the susceptible growth period of the host in relation to disease development, the plants were raised from surface sterilized healthy seeds of *Dolichos lablab*, L. variety "Kalyanpur Type-1" in earthen pots of 30.0 cm. size filled with sterilized soil. These pots were kept in glasshouse. Four plants per pot were maintained and three replications, were kept for each age. For maintaining the plants of different growth stages, sowing was done at the interval of 10 days.

These plants, were inoculated with the mycelial-cum-spore suspension of 10 days old culture of the pathogen in the glass house simultaneously after 10 days of last sowing. After inoculation the plants were incubated for 48 hours in

moist chamber by covering the plants with polythene bags. Data on disease intensity was recorded in per cent leaf area covered after 15 days of inoculation.

13. EFFECT OF CLIMATIC CONDITIONS AFFECTING THE DEVELOPMENT OF DISEASE -

To determine the effects of atmospheric temperature, relative humidity and rainfall on disease development affecting the development of disease in highly susceptible variety "Kalyanpur Type-1" was sown in the field during the crop seasons of 2001 and 2002. Thirty days old plants were inoculated at fortnight intervals in both the years by spraying spore suspension. The field was watered periodically, whenever necessary. The data on per cent disease intensity was recorded fortnightly in per cent leaf area covered after 15 days of inoculation at seven days interval. The weather data was collected during the crop period from meteorological Department and were correlated with the disease development. Ten plants were randomly selected and data were recorded on them, were averaged for each date for precision.

14. DETERMINATION OF BIOCHEMICAL CONSTITUENTS -

Studies, were taken to find out the significance of different biochemical parameters in disease resistance/susceptibility against leaf spot of Dolichos bean. For this purpose the most susceptible variety "Kalyanpur Type-1" was used.

(I) COLLECTION OF LEAF SAMPLES -

The experiments were conducted in 2002 in pot culture. Three sets of plants of variety "Kalyanpur Type-1" were raised in 30.0 cm. pots and four plants per pot were maintained for inoculating them. In one set the plants were inoculated after the age of 25 days and in another set after 50 days with mycelial-cum-spore suspension of the pathogen and kept in moist chamber for 48 hours and then transferred in the open and the leaves were collected for analysis after 15 days of inoculation. In third set variety "Kalyanpur Type-1" was kept in glasshouse conditions, without inoculation of pathogen working as control. The plants were well watered regularly in order to maintain sufficient moisture. Healthy and diseased leaves were collected from the inoculated plants at the crop age of 40 and seventy days.

II. CHEMICAL ANALYSIS OF LEAF SAMPLE -

The healthy and diseased leaves of susceptible variety "Kalyanpur Type-1",

were firstly dried for two days in shade in order to remove excessive moisture. Afterwards these leaves were oven dried at 60°C for 24 hours by converting into powder form by grinding used for analysis of polyphenols, sugars, nitrogen, phosphorus, potash and sulphur after passing through 20 mesh sieve. The surface wax and chlorophyll content, were estimated from the fresh leaes.

(A) Wax content -

The surface wax content, was analysed by the method of John *et al.* (1978) using Chloroform a solvent. The result is expressed as mg./g. fresh weight of leaves.

(B) CHLOROPHYLL -

Total chlorophyll (a and b), was determined according to the technique described by Arnon (1949). The results are expressed in mg./g. fresh weight of leaves. Chlorophyll-a and chlorophyll-b contents, were determined by the help of following formula.

$$\text{Chlorophyll-a} = (12.7 \times \text{O.D.} 663 - 2.69 \times \text{O.D.} 645) \times \frac{V}{100} \times W.$$

$$\text{Chlorophyll - b} = (22.90 \times \text{O.D.} 645 - 4.68 \times \text{O.D.} 663) \times \frac{V}{100} \times w$$

where, total chlorophyll content = Chlorophyll - a + Chlorophyll - b

V = Final volume of extract in 80.0 per cent acetone (25.0 ml.)

W = Fresh weight of leaf sample in gm. (0.50 mg.)

O.D. 663 and O.D. 645 optical density at 663 m. and 645 m. respectively spectronic 20 colorimeter.

(C) TOTAL POLYPHENOLS :-

The polyphenol contents, were extracted from the dry leaves using 80.0 per cent alcohol (methanol) and estimated as tannic acid equivalent according to Folin-Denis technique suggested by Swain and Hills (1959). The results are expressed as mg./g. dry weight of leaves.

(D) SUGAR CONTENTS -

Reducing and non-reducing sugars, were analysed by Ferrieyanid technique as given in AOAC (1970). The results are expressed as mg./g. on dry

weight basis.

(E) NITROGEN CONTENT -

Nitrogen content in dry leaves was determined by Kjeldahl technique as described by AOAC (1970).

(F) PHOSPHORUS CONTENT -

The phosphorus content in dry leaves was estimated by Colorimetric method according to the technique suggested by Jackson (1973).

(G) POTASSIUM CONTENT -

The potassium content in the dry leaves was determined according to the photometric method as described by Jackson (1973).

(H) SULPHUR CONTENT -

The sulphur content was determined according to technique suggested by Chesnin and Yien (1950) by use of Spectronic-20 colorimeter.

15. MODE OF PERPETUATION AND SPREAD OF PATHOGEN (PRIMARY AND SECONDARY SPREAD OF DISEASE) -

In order to study the survival of pathogen (infection, perpetuation and spread of disease) the following experiments were conducted under laboratory and field conditions :-

(A) MODE OF PRIMARY SPREAD OF DISEASES -

1. ROLE OF SOIL -

For ascertaining the role of soil in perpetuation of the pathogen, the soil taken from field having natural infection of leaf spot of the crop, was mixed with sterilized soil and stored at room temperature $25\pm 1^{\circ}\text{C}$ in polythene bags. Monthly isolations, were done from these soils to ascertain the perpetuation of the pathogen.

For determination of role of infested soil as a primary source of infection the pots were filled with naturally infested soil and artificially infested sterilized soil. Twenty five sterilized healthy seeds obtained from healthy plants were sown in each pot. The pots filled with sterilized soil sown with surface sterilized seeds served as control. Each treatment was replicated three times. These pots were kept in the glass house to avoid aerial infection. The regular

examination of growing plants, revealed the disease development.

2. ROLE OF PLANT DEBRIS -

In order to study the survival and perpetuation of the pathogen the following experiments were conducted under laboratory and field conditions :-

(A) UNDER LABORATORY CONDITIONS -

The affected leaves from naturally infected *Dolichos* bean plants were collected in last week of 2002 and wrapped in a tissue paper after drying between blotting papers and stored in the laboratory for one year till the next crop season at $25\pm 1^{\circ}\text{C}$. Periodical isolations were made from this material for 12 months to observe the viability of the pathogen alongwith regular inoculation experiments to establish the pathogenicity.

(B) UNDER FIELD CONDITIONS -

Diseased plant debris, wrapped in tissue paper, was buried in the soil at a depth of 10.0 cm. and kept in the open ($4-48^{\circ}\text{C}$). Regular isolations and pathogenicity tests, were carried for one year at monthly intervals to study the longevity of pathogen.

With a view to study the role of diseased plant debris as primary source of infection, two sets of experiments were conducted. In one set three earthen pots of 30.0 cm. diameter, were filled with sterilized soil and heavily infected diseased plant debris stored at room temperature ($25\pm 1^{\circ}\text{C}$). In another set the diseased plant debris was buried in the pot soil and kept in open at $5^{\circ}\text{C} - 45^{\circ}\text{C}$, was thoroughly mixed with sterilized soil and filled in pots in three replications. In all these pots the surface sterilized healthy seeds were sown at the rate of 5 seeds of *Dolichos* bean variety "Kalyanpur Type-1" obtained from healthy plants. The pots filled with sterilized soil and sown with surface sterilized seeds served as control. The pots were kept in glass house chamber to avoid the aerial contamination. The inoculated plants were regularly observed for disease development.

3. ROLE OF SEED :-

For the study of determination of role of seeds as a carrier of the pathogen, surface sterilized healthy seeds were mixed with the spore suspension of the pathogen and sterilized seeds obtained from diseased plants, were stored in paper envelopes at room temperature $25\pm 1^{\circ}\text{C}$. Monthly isolations were made

from both healthy and affected seeds in petridishes with Potato dextrose agar (P.D.A.) medium to establish the survival and association of pathogen with the seeds. These petridishes were then incubated at $25\pm 1^{\circ}\text{C}$ and examined for the presence of pathogen.

In order to study the role of seeds as primary source of infection of disease, the seeds collected from naturally affected plants as well as mixed with the spore suspension of the pathogen, were sown in pots filled with sterilized soils. Twenty seeds were sown in each pot with three replications. Surface sterilized seeds in pots having sterilized soil served as control. The source of aerial infection, was checked by keeping the pots in glasshouse. The observations on disease appearance, were recorded regularly.

(B) MODE OF SECONDARY SPREAD OF DISEASE -

The secondary spread of disease was ascertained by air borne inoculum by raising the healthy plants in glasshouse by sowing 10 surface sterilized seeds per pot filled with sterilized soil. One set of these pots was covered with muslin cloth in order to avoid aerial contamination, while other similar set was kept as exposed to free contact of spores of the pathogen present in air. The diseased plants raised in pots were transferred in the vicinity of healthy plants to study the role of air in the secondary spread of disease. The appearance of the disease on the healthy plants, was observed regularly.

16. VARIETAL SCREENING FOR DISEASE RESISTANCE :-

The experiments were conducted to find out the possibility of combating leaf spot of *Dolichos* bean through varieties/cultures.

(A) UNDER NATURAL CONDITIONS -

The germplasm of *Dolichos* bean (*Dolichos lablab*, L.), was screened against the pathogen under natural conditions. Germplasms/cultures of *Dolichos* bean variety "Kalyanpur Type-1", were sown in randomized block design with three replications in a paired row plots of 2.50 meter length. The fertilizer dose of N80-P60 and K-40 per hectare was applied. The plots were irrigated from time to time to maintain sufficient moisture. To test the germplasm for disease resistance one interior row after every fifth line of the test germplasm was kept in order to built up good inoculum potential. The disease intensity was recorded on the basis of percent leaf area affected at the time of harvest of the crop by randomly selecting 50 leaves from each replication according to the technique

suggested by Chopra and Sharma (1976). The varieties and cultures were graded into six grades/categories as detailed in Table-V given below -

Table V

Particulars of different categories of Disease resistance on the germplasms inoculated by the fungus, *Alternaria alternata* (Fries.), Keissler causing leaf spot of Dolichos bean (*Dolichos lablab*, L.)

S.No.	Grade	Reaction	Rating	Details of Infection
1.	0	Nil	—	No infection
2.	1	Resistant	+	Up to 50.0 % leaf area affected
3.	2	Moderately resistant	+	5-10% leaf area affected
4.	3	Moderately susceptible	+++	11-20% leaf area affected
5.	4	Susceptible	++++	20-30% leaf area affected
6.	5	Highly Susceptible	+++++	Above 30% leaf area affected

(B) UNDER ARTIFICIAL CONDITIONS -

The germplasms found resistant and moderately resistant under natural conditions were further tested under conditions of artificial epiphytotic of the disease. Varieties were screened in pots under artificial inoculation for their reaction to pathogen. For this study seeds of each Dolichos bean variety/culture were sown in 30.0 cm. earthen pots in three replications. Four plants per pot were maintained. After age of 45 days of crop the plants were inoculated by spraying with an automizer with the spore-cum- mycelial suspension of 10 days old culture of pathogen, *Alternaria alternata* (Fries.) Keissler. In order to maintain proper moisture the pots were watered before inoculation and were kept in humid chamber for 48 hours and then transferred to open. Sufficient moisture was maintained by spraying water outside the humid chamber. Observations on the disease severity was recorded after 15 days of inoculation

as given earlier in natural conditions and germplasms were categorised in terms of per cent leaf area according to the technique suggested by Chopra and Sharma (1976) given in Table V.

17. HOST RANGE OF THE PATHOGEN -

The species of genus *Alternaria* is well known to parasitize a wide variety of hosts viz., crop plants, vegetable plants, fruit yielding plants, ornamental plants etc. The determination of the ability of test fungus, *A. alternata* (Fries.), Keissler to infect hosts other than Dolichos bean (*Dolichos lablab*, L.) was ascertained by conducting experiments. For this plants belonging to different genera of different families were studied. The seeds were sown in earthen pots filled with sterilized soil. Under each treatment four plants were taken and replicated three times. After attaining the age of 45-60 days the plants were inoculated with spore-cum-mycelial suspension of the pathogen. The pots containing test plants were kept in moist chamber for 24 hours prior and 48 hours after inoculation to facilitate penetration of pathogen and establishment of infection. In each treatment one pot out of the five plants was not inoculated and served as control. The pots were placed at room temperature $25\pm 1^{\circ}\text{C}$ and watched regularly for the appearance of disease. The observations were recorded after a fortnight of inoculation.

18. CHEMICAL CONTROL OF THE DISEASE -

In the present investigation different seed dressing and spray fungitoxics were tested for the control of leaf spot of Dolichos bean belonging to different groups viz., Benzene, Copper compound, Dithiocarbamate, Heterocyclic nitrogenous compounds, Miscellaneous organomercurials, Quinone, Systemic and Sulphur compounds as well as an antibiotic. Prior to perform these studies the efficacy of various fungitoxics and an antibiotic were ascertained in bioassay test against the pathogen and those found effective were used in further experiments in pots/fields for control of the disease. The following methods were employed in these studies :-

(A) SCREENING OF FUNGITOXICANTS AGAINST THE PATHOGEN IN VITRO :-

Twenty five fungitoxics and an antibiotic detailed in Table VI were used for their relative efficacy against the pathogen in laboratory in inhibiting the growth of pathogen in culture according to the "Food Poison", technique suggested by Schmitz (1930). The relative efficacy was determined in *vitro* by

measuring colony diameter, weighing dry weight, recording increase in lesion development, size and spore germination in culture.

TABLE VI

Fungitoxicants and an antibiotic used against the pathogen, *Alternaria alternata* (Fries.) Keissler, for the study of their efficacy.

Sl. No.	Group	Name of Fungitoxicants and Antibiotic
A.	Antibiotic	1. Aureofungin (N-Methyl- p-aminoacetophenone mycos amineheptone)
B.	Benzene Halogenated organic compound	1. Brassicol. (75 w.p.) 1, 2, 3, 4, 5, Pentachloronitrobenzene (P.C.N.B.)
C.	Dithiocarbamate compound	1. Dithane M-45 (78 % Zn, Fe, Mn complex of ethylene bisdithio- carbamate) 2. Dithane Z-78 (75% ethylene bisdithiocarbamate) 3. Karathane (EC-D-1-methyl, 2 heptyl, 4, 6 dinitrophenyl crotonate) 4. Duter (50% W.P. Ferric dimethyl dithiocarbamate) 5. Ferbam (75% W.P. Ferric dimethylcarbamate) 6. Thiram (Tetramethyl thiruram disulphide) 7. Ziram (75% W.P., Zinc dimethyl dithiocarbamate)

- | | | |
|----|-----------------------------------|--|
| D. | Heterocyclic nitrogenous compound | <ol style="list-style-type: none"> 1. Captan (50% N-trichloro methyl thiomercapto-4 cylohexane, 1, 2, dicarboximide) 2. Foltaf 80 W. (Captafol, Cix-N-1, 1, 2, 2 tetrachloromethyl thio sulenmya-4-cyclohexene 1, 2 dicarboximide) |
| E. | Inorganic Copper Compound | <ol style="list-style-type: none"> 1. Blitox-50 (50% Copper oxychloride) |
| F. | Inorganic Sulphur Compound | <ol style="list-style-type: none"> 1. Suflex (80% W.P. micronized Sulphur) 2. Hexaferb (Ferric dimethyl carbamate) |
| G. | Organomercury Compound | <ol style="list-style-type: none"> 1. Agrosan G.N. (Phenylmercury acetate ethyl mercury chloride) 2. Ceresan Dry (Phenyl mercury acetate, 1% mercury) 3. Emisan-6 (Methoxyethyl mercuric chloride, 6% mercury) |
| H. | Quinone Compound | <ol style="list-style-type: none"> 1. Dichlone (2, 3 dichloro, 1, 4 napthaquinone) 2. Spergon (Tetrachlor-b-benzoquinone) |
| I. | Systemic | <ol style="list-style-type: none"> 1. Bavistin (2-metnoxycarbomyl benzimidazole) 2. Calixin (N-tridecyl-2-6 dimethyl morpholine) 3. Pancotine (Guanidated, '9-aza-1, 17-diaminohepta diecane acetate salt). |

4. Ridomil (Methyl D-LN-12, 6-dimethyl phenyl 7-N-12 methoxy acetyl alaninate)
5. Vitavax (75%, 5, 6, Dihydro-2 Methyl 1, 40-xanthin-3-Carboxanilids DMOC).

J. Miscellaneous

1. Benlate (Methyl-1, Butyl carbomyl - 1 Benzimidazole carbamate)
2. Kavach (750/0 Tetrachloro isophthalonitrite).

(i) RADIAL GROWTH OF FUNGAL COLONY -

The requisite quantities of the fungitoxicants and an antibiotic, were incorporated in 2.0 per cent sterilized Potato dextrose agar medium and shaken well to make it homogenous prior to pouring in petridishes. The fungitoxicants impregnated medium, was then poured in sterilized petridishes in three replications for each treatment and allowed to solidify. Afterwards these petridishes were inoculated with 5.0 mm. circular agar discs of fungal inoculum cut from the edge of growing colony from 10 days old culture. The inoculum, was placed in centre of each petridish in such a way so that fungus may come in direct contact with the medium. The medium without any fungitoxicant poured and inoculated served as control. The petridishes were incubated at $25 \pm 1^\circ\text{C}$ for 10 days. The efficacy of various fungitoxicants was assessed by measuring the radial growth of fungal colony in mm.

The percent inhibition over control was calculated by the formula given by Bliss (1934) :-

$$\text{Percent inhibition over control} = \frac{C - T}{C} \times 100, \text{ where}$$

C = Growth of the fungus in control.

T = Growth of fungus in treatment.

The fungitoxicants, which were found effective in laboratory evaluation,

were further tried in seed treatment and finally as spray fungitoxicant in the field.

(ii) HYPHAL DRY WEIGHT :

On Czapek's-dox liquid medium dispensed into 250.0 ml. Erlenmeyer's flasks were autoclaved. Aliquots of fungicides prepared in sterilized water, were mixed in cooled molten medium to give final concentration of active ingredients at 0.05, 0.10, 0.15, 0.20, 0.40 and 0.60 in 100 ml. of total volume in each flask. Sterilized water in equal volume was added in control sets. Three replicates were used for each treatment. Flasks were inoculated with 5.0 mm. circular fungal discs cut from the edge of colony of test fungus. The flasks were vigorously shaken and incubated for 10 days at $25\pm 1^{\circ}\text{C}$. After incubation period mycelia were harvested over a pre-weighed, Whatman's filter paper No. 42, washed and then dried to a constant weight.

(iii) LESION MEASUREMENT -

The fungitoxicants, were assessed for their protective action by observing the inhibition on the development of lesions produced by *Alternaria alternata* (Fries.), Keissler causing leaf spot of *Dolichos* bean. Fresh healthy leaves of same age having no diseased symptoms were excised and collected in polythene bags. Fungitoxicants in dilutions of 0.05, 0.10, 0.15, 0.20, 0.25, 0.30, 0.35 and 0.40 per cent were inoculated by spraying with an automizer at the point of injury made. Fungitoxicants were allowed to soak and dry up. Afterwards leaves were inoculated with one drop of 0.001 ml. of spore suspension at four points of leaf lamina and were kept in petridishes in moist chamber over surface sterilized bent glass rods to avoid direct contact with moist filter paper. All sets were incubated at room temperature.

The brown spot development of disease causing leaf spot of *Alternaria* was evaluated after 10 days on the leaves by use of scale 0-5. Six inoculation sets of each leaf were rated and rating from three leaf discs were averaged. Sterilized distilled water was used in place of fungitoxicants sprayed in controlled sets. The different type of symptoms produced due to pathogenic attack of *Alternaria alternata* on *Dolichos lablab*, L. as leaf spot are presented in following Table VII.

TABLE VII.

Different kinds of categories for the study of pathogenic symptoms produced by pathogen, *Alternaria alternata* (Fries.), Keissler causing leaf spot of Dolichos bean (*Dolichos lablab*, L.)

S.No.	Grade	Reaction and Description of Symptoms	Rating
1.	0	Nil	—
2.	1	Light yellow lesions	LY
3.	2	Yellowish brown lesions	YB
4.	3	Light brown lesions	LB
5.	4	Extensive brown lesions	EB
6.	5	Phytotoxic effects	PE

(iv) SPORE GERMINATION TEST -

For the study of spore germination test a suspension of conidia of the test fungus, *Alternaria alternata* (Fries.), Keissler was prepared. Fungicides and an antibiotic in dilutions of 0.0002, 0.0004 and 0.0006 per cent were prepared in sterilized water (one drop approximately 0.0001 ml.), were separately inoculated by way of dispensing in glass cavity slides in three replications. One drop of spore suspension containing fungicides was thoroughly mixed and poured in the cavity of slide. The one drop suspension contains nearly 100 spores previously, standardised by a Haemaocytometer. The cavity slides were kept in petridishes lined with moist filter paper for 24 hours and incubated for 10 days at room temperature, $25\pm 1^{\circ}\text{C}$. At the end of incubation period the spores were mounted in lactophenol and observations of spore germination viz., number of spores germinated, length and number of germ tubes were recorded. For control spore suspension was dispersed in cavity slides containing sterilized distilled water.

(B) EFFICACY OF FUNGICIDES IN SEED TREATMENT

The efficacy of Eleven fungicides viz., Agrosan G.N. Aureofengin, Captan Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Pancotine, Thiram and Vitavax were evaluated for eliminating the seed borne mycoflora by Standard blotter method.

The naturally infected seeds of high seleptible variety "Kalyanpur Type-1"

due to presence of infection of *Alternaria alternata*, were treated with fungicides and an antibiotic by mixing their required quantities and shaking the seeds in plugged conical flasks for 15 minutes. The seeds treated with each fungitoxicant, were transferred to sterilized moist blotter paper at the rate of 25 seeds per blotter and each treatment was replicated three times. Plates treated with untreated seeds were also maintained in another set, which served as control. Observations were recorded on the germination of seeds and appearance of fungal growth on seed and seedlings were recorded after 10 days of incubation at $25\pm 1^{\circ}\text{C}$. Percentage of seed germination and seed infection were calculated on the basis of total number of seeds selected.

Further to examine the efficacy of various seed dressing fungitoxicants and an antibiotic under field conditions in improving seed germination and seedling infection, fifty seeds were treated with each fungitoxicant were sown in 30.0 cm. earthen pots and replicated three times. Pots sown with untreated seeds served as control. Observations on seed germination and seedling infection were noted after 10 days of sowing.

(C) EVALUATION OF FUNGICIDES IN THE FIELD

The fungitoxicants found effective under laboratory conditions, were examined for the control of disease under field conditions in pot experiments in field trials of 2001 and 2002.

(i) EVALUATION OF FUNGITOXICANTS IN POT EXPERIMENTS :-

The plants of high susceptible variety "Kalyanpur Type-1" of Dolichos bean was raised in 30.0 cm. earthen pots and five plants per pot were maintained and replicated three times.

After attaining the age of 45 days the plants were inoculated with mycelial-cum-spore suspension of the pathogen. Fungitoxicants and an antibiotic were sprayed after 48 hours after inoculation at an interval of 10 days with two subsequent sprays. The potted plants sprayed with sterilized water served as control.

Observations on disease intensity were noted on the basis of percent leaf area affected for 10 days of last spraying. Yield data were also recorded per plant after harvest of the crop in each treatment.

(ii) EVALUATION OF FUNGITOXICANTS IN FIELD TRIALS :-

Field trials were conducted with the same fungitoxicants and an antibiotic as used in pot experiments by sowing most susceptible variety "Kalyanpur Type-1" in the seasons of 2001 and 2002 for control of disease. The soil was also prepared and recommended doses of fertilizers and manures were applied. The experiment was conducted in randomized block design with three replications using plot size of 2.0 m. × 5.0 cm. and spacing between the rows and plants were 60.0 cm. and 30.0 cm. respectively. Artificial epiphytotics of the disease were created by inoculation of 45 days aged plants by spraying with 10 days old mycelial-cum-spore suspension of the test pathogen, *Alternaria alternata* and the plots were irrigated from time to time to maintain proper moisture.

The first spray of fungitoxicant and an antibiotic was given after 48 hours of inoculation followed by two applications of an interval of ten days. The control plots were sprayed with water only.

Final observations on disease intensity were noted after 15 days of last spray of fungitoxicants on the basis of percent leaf area affected. Yield per plot was recorded after harvesting the crop.

Per cent disease control and per cent deviation in yield over control was calculated by the following formulae :-

Per cent disease control =

$$\frac{\text{Disease intensity in control} - \text{Disease intensity in treatment}}{\text{Disease intensity in control}} \times 100$$

Per cent deviation and yield due to treatment =

$$\frac{\text{yield in treatment} - \text{yield in control}}{\text{yield in control}} \times 100$$

□□□

Chapter - IV

EXPERIMENTAL FINDINGS

EXPERIMENTAL FINDINGS

SURVEY FOR THE STUDY OF PREVALENCE AND SEVERITY OF DISEASE -

In order to study the prevalence and severity of leaf spot of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, under natural conditions, a survey was conducted at the various Research Stations of Uttar Pradesh situated at different places during Kharif season of the crop in the years 2001 and 2002. The results of surveys are detailed in Tables VII and VIII and Figures 1 and 2.

TABLE - VII

Prevalence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, at different Research Stations of U.P. in the Kharif crop season of the year 2001.

S. No.	Name of Research Station	Location	Germplasm	Crop season
1.	Agriculture Science Centre	Ganiwa, Chitrakoot	Culture-7301	+
2.	Charyar Farm	Bulandshahar	HD-4	+
3.	Crop Research Farm	Etawah	Culture-6801	+
4.	Regional Research Station	Saini, Allahabad	Culture-7015	+
5.	Crop Research Farm	Saraimira, Farrukhabad	Culture-8101	+
6.	Crop Research Farm	Baraur, Farrukhabad	JD-L-85	+
7.	Crop Research Farm	Araul, Kanpur	Culture-8403	+
8.	Crop Research Farm	Deegh, Kanpur	Culture-9104	-
9.	Crop Research Farm	Modipuram Area, Meerut	Culture-9113	+

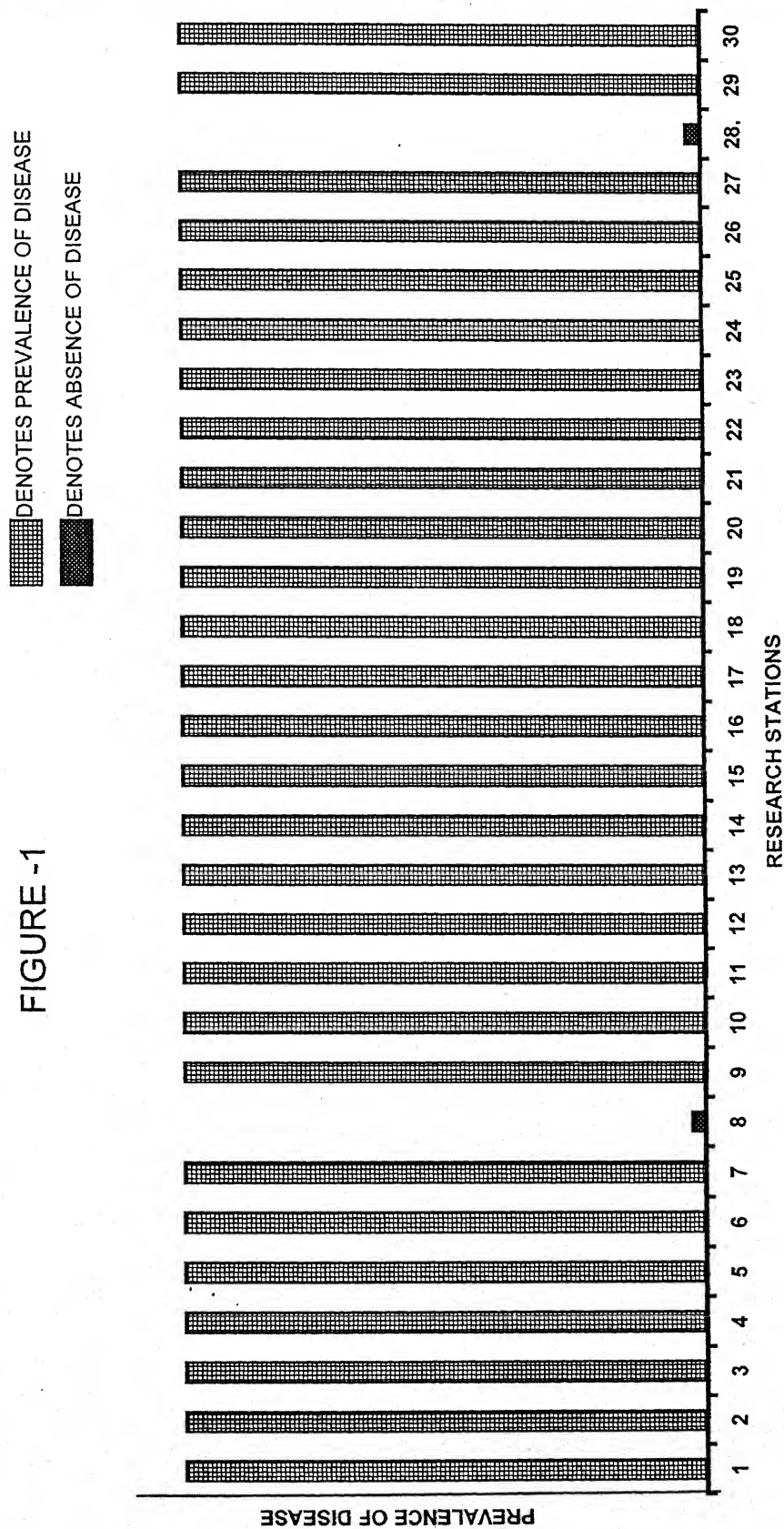
10.	Crop Research Centre	Mauranipur, Jhansi	Rajani	+
11.	C.S. Azad University Research Farm	Belatal, Mahoba	Culture-9118	+
12.	Directorate of Vegetable Research Farm	Varanasi	Todi-125-136	+
13.	G.P. Pant University Farm	Pant Nagar, Nainital	Culture-7210	+
14.	Govt. Agriculture Centre	Atarra, Banda	Culture-7710	+
15.	Govt. Agriculture Centre	Bansi, Banda	Culture-8005	+
16.	Govt. Agriculture Centre	Khaptiha, Banda	Culture-9102	+
17.	Govt. Agriculture Research Centre	Bharari, Jhansi	Kamgranj Selection - 2	+
18.	Govt. Agriculture Centre	Bagi, Jalaun	HD-93	+
19.	Groundnut Research Station	Mainpuri	Pusa Early Prolific	+
20.	N.D. University	Kumarganj, Faizabad	Culture-9109	+
21.	Oil Seed Research Farm	Kalyanpur, Kanpur	Culture-7604	+
22.	Regional Research Centre	Amroha, Jhansi	Culture-7708	+
23.	Regional Research Station	Uttaripura Kanpur Dehat	JDL-79	+
24.	Regional Research Centre	Hardoi	HD-66	+
25.	Regional Research Centre	Varanasi	DB-1	+
26.	Regional Research Station	Madhurikund, Mathura	Goya	+
27.	Regional Research Station	Dilip Nagar, Kanpur	Hatikan	+

FIGURE - 1

Prevalence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. in Kharif crop season of the year, 2001.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE -1



PREVALENCE OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE KHARIF CROP SEASON OF THE YEAR 2001

28.	Regional Agriculture Demonstration Centre	Meerut	DPL-1	-
29.	Vegetable Research Farm	C.S. Azad University of Agriculture and Technology, Kanpur	Kalyanpur Type - 1	+
30.	Vegetable Research Farm	Kalyanpur, Kanpur	Akra Jai	+

(+) = Denotes Prevalence of disease.

(-) = Denotes Absence of disease.

TABLE - VIII

Prevalence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, at different Research Stations of U.P. in the Kharif crop season of the year 2002.

S. No.	Name of Research Station	Location	Germplasm	Crop season
1.	Agriculture Science Centre	Ganiwa, Chitrakoot	Culture-7301	+
2.	Charyar Farm	Bulandshahar	HD-4	+
3.	Crop Research Farm	Etawah	Culture-6801	+
4.	Regional Research Station	Saini, Allahabad	Culture-7015	+
5.	Crop Research Farm	Saraimira, Farrukhabad	Culture-8101	+
6.	Crop Research Farm	Baraur, Farrukhabad	JD-L-85	+
7.	Crop Research Farm	Araul, Kanpur	Culture-8403	+
8.	Crop Research Farm	Deegh, Kanpur	Culture-9104	+

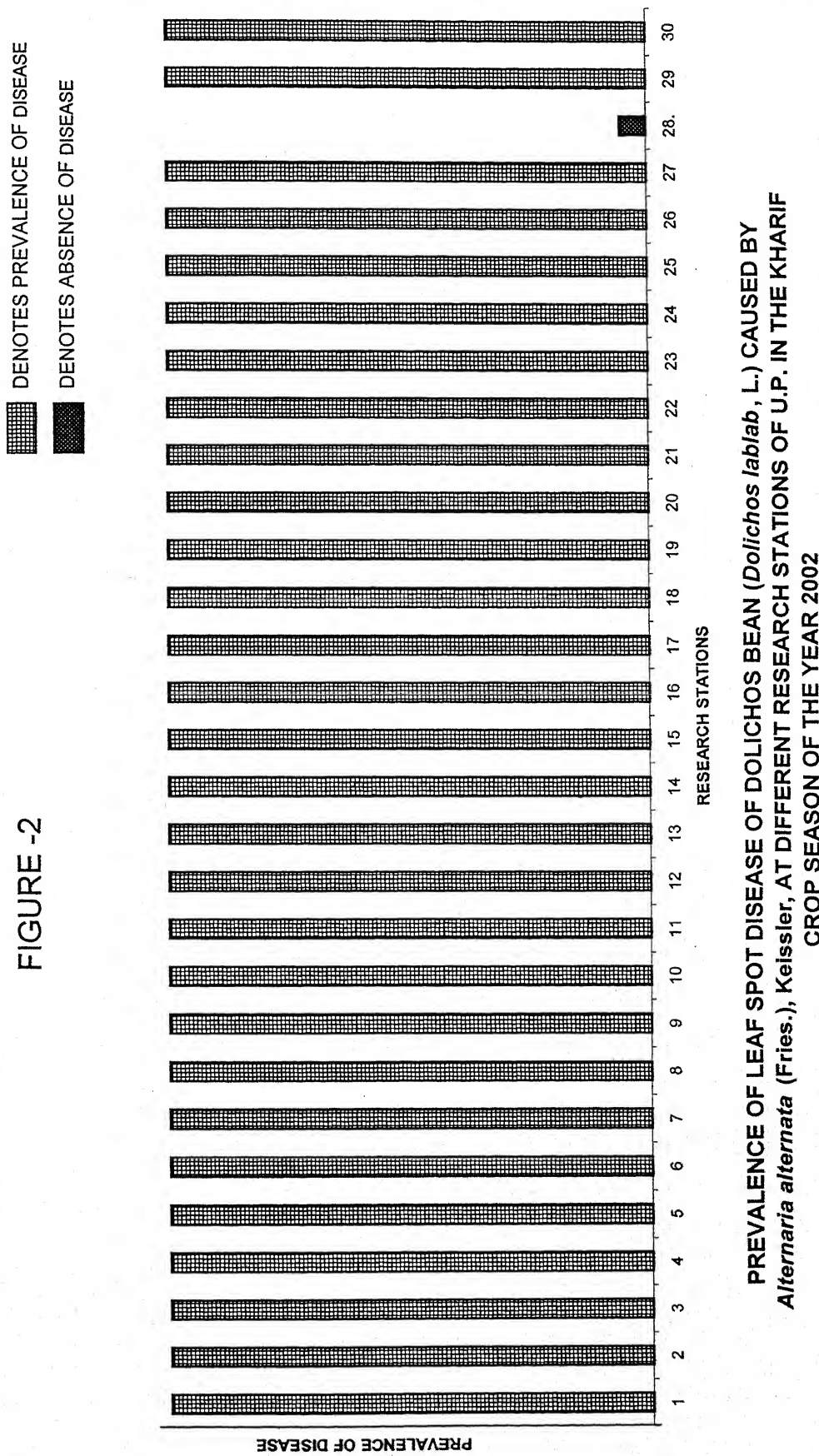
9.	Crop Research Farm	Modipuram Area, Meerut	Culture-9113	+
10.	Crop Research Centre	Mauranipur, Jhansi	Rajani	+
11.	C.S. Azad University Research Farm	Belatal, Mahoba	Culture-9118	+
12.	Directorate of Vegetable Research Farm	Varanasi	Todi-125-136	+
13.	G.P. Pant University Farm	Pant Nagar, Nainital	Culture-7210	+
14.	Govt. Agriculture Centre	Atarra, Banda	Culture-7710	+
15.	Govt. Agriculture Centre	Bansi, Banda	Culture-8005	+
16.	Govt. Agriculture Centre	Khaptiha, Banda	Culture-9102	+
17.	Govt. Agriculture Research Centre	Bharari, Jhansi	Kamgranj Selection - 2	+
18.	Govt. Agriculture Centre	Bagi, Jalaun	HD-93	+
19.	Groundnut Research Station	Mainpuri	Pusa Early Prolific	+
20.	N.D. University	Kumarganj, Faizabad	Culture-9109	+
21.	Oil Seed Research Farm	Kalyanpur, Kanpur	Culture-7604	+
22.	Regional Research Centre	Amroha, Jhansi	Culture-7708	+
23.	Regional Research Station	Uttaripura Kanpur Dehat	JDL-79	+
24.	Regional Research Centre	Hardoi	HD-66	+
25.	Regional Research Centre	Varanasi	DB-1	+

FIGURE - 2

Prevalence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. in Kharif crop season of the year, 2002.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE -2



26.	Regional Research Station	Madhurikund, Mathura	Goya	+
27.	Regional Research Station	Dilip Nagar, Kanpur	Hatikan	+
28.	Regional Agriculture Demonstration Centre	Meerut	DPL-1	-
29.	Vegetable Research Farm	C.S. Azad University of Agriculture and Technology, Kanpur	Kalyanpur Type - 1	+
30.	Vegetable Research Farm	Kalyanpur, Kanpur	Akra Jai	+

(+) = Denotes Prevalence of disease.

(-) = Denotes Absence of disease.

DISEASE INCIDENCE -

The incidence of the disease was recorded in the field by method described under "Material and Method". The data collected is presented in Tables IX and X and Figures 3, 4, 5 and 6.

TABLE - IX

Prevalence and incidence of Leaf spot Disease of *Dolichos bean* (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, at different Research Stations of U.P. in the 2001 in Kharif season.

S. No.	Location	Number of Plants in the field	Number of affected plants in the field	Average Disease incidence in %
1.	Agriculture Science Centre, Ganiwa, Chitrakoot	97	13	13.48
2.	Charyar Farm, Bulandshahar	104	21	20.19

3.	Crop Research Farm, Etawah	96	22	22.91
4.	Regional Research Station, Saini, Allahabad	105	16	17.14
5.	Crop Research Farm Saraimira, Farrukhabad	123	26	23.57
6.	Crop Research Farm Baraur, Farrukhabad	133	38	28.59
7.	Crop Research Farm Araul, Kanpur	132	27	20.45
8.	Crop Research Farm Deegh, Kanpur	—	—	—
9.	Crop Research Farm Modipuram Area, Meerut	110	31	28.18
10.	Crop Research Centre Mauranipur, Jhansi	102	23	22.54
11.	C.S. Azad University Research Farm, Belatal, Mahoba	92	16	17.39
12.	Directorate of Vegetable Research Farm, Varanasi	67	14	20.89
13.	G.P. Pant University Farm, Nainital	89	22	24.71
14.	Government Agriculture Centre, Atarra, Banda	145	38	26.20
15.	Government Agriculture Centre, Bansi, Banda	127	36	28.34
16.	Government Agriculture, Centre, Khaptiha, Banda	65	29	15.38
17.	Government Agriculture Research Centre, Bharari, Jhansi	96	16	16.66

18.	Government Agriculture Centre, Bagi, Jalaun	118	37	38.50
19.	Groundnut Research Station, Mainpuri	125	27	21.60
20.	N.D. University Kumarganj, Faizabad	132	33	25.0
21.	Oil Seed Research Farm, Kalyanpur, Kanpur	84	18	21.42
22.	Regional Research Centre, Amroha, Jhansi	75	14	18.66
23.	Regional Research Station, Uttaripura, Kanpur Dehat	82	17	21.95
24.	Regional Research Centre, Hardoi	108	26	24.07
25.	Regional Research Centre, Varanasi	93	19	20.43
26.	Regional Research Station, Madhurikund, Mathura	78	17	21.79
27.	Regional Research Station, Dilip Nagar, Kanpur	89	21	23.59
28.	Regional Agriculture Demonstration Centre, Meerut	—	—	—
29.	Vegetable Research Farm Chandra Shekhar Azad University of Agriculture and Technology, Kanpur	152	48	31.57
30.	Vegetable Research Farm Kalyanpur, Kanpur	118	24	20.33

(-) = Denotes absence of Disease.

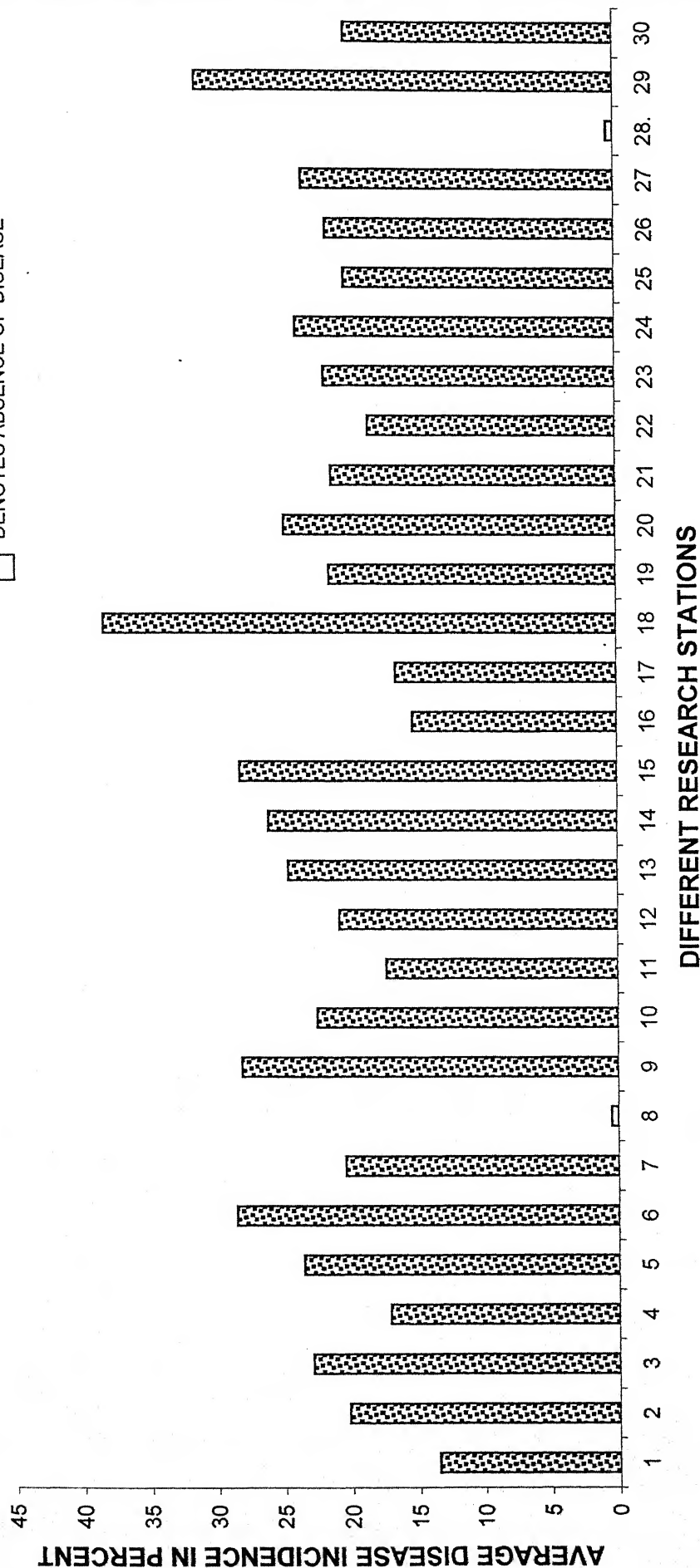
FIGURE - 3

Prevalence and incidence of Leaf spot Disease of *Dolichos bean* (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. of 2001 in Kharif season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE -3

 DENOTES PREVALENCE AND INCIDENCE OF DISEASE
 DENOTES ABSENCE OF DISEASE



DIFFERENT RESEARCH STATIONS

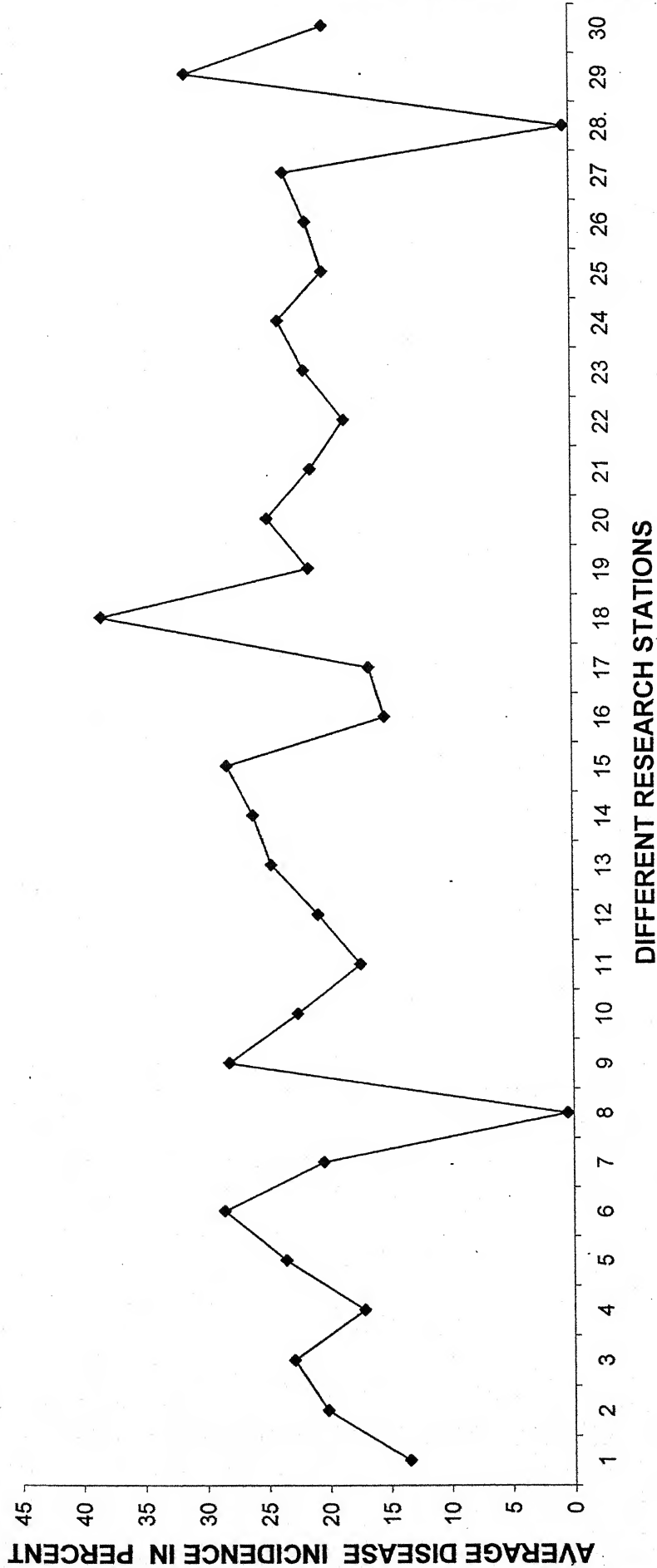
PREVALENCE AND INCIDENCE OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2001 IN KHARIF SEASON

FIGURE -4

Prevalence and incidence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. In the year, 2001 in Kharif crop season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Farm Centre Mauraipur, Jhansi.
11. C.S.Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jaluan.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumarganj, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagar, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE-4



PREVALENCE AND INCIDENCE OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2001 IN KHARIF SEASON

TABLE-X

Prevalence and incidence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, at different Research Stations of U.P. in the year 2002 in Kharif season.

S. No.	Location	Number of Plants in the field	Number of affected plants in the field	Average Disease incidence in %
1.	Agriculture Science Centre, Ganiwa, Chitrakoot	195	20	10.25
2.	Charyar Farm, Bulandshahar	110	26	23.63
3.	Crop Research Farm, Etawah	87	12	13.79
4.	Regional Research Station, Saini, Allahabad	116	20	17.24
5.	Crop Research Farm Saraimira, Farrukhabad	133	32	24.06
6.	Crop Research Farm Baraur, Farrukhabad	79	16	20.25
7.	Crop Research Farm Araul, Kanpur	119	30	25.21
8.	Crop Research Farm Deegh, Kanpur	137	39	28.46
9.	Crop Research Farm Modipuram Area, Meerut	106	31	29.24
10.	Crop Research Centre Mauranipur, Jhansi	93	26	27.95
11.	C.S. Azad University Research Farm, Belatal, Mahoba	109	15	13.76
12.	Directorate of Vegetable Research Farm, Varanasi	142	15	17.60

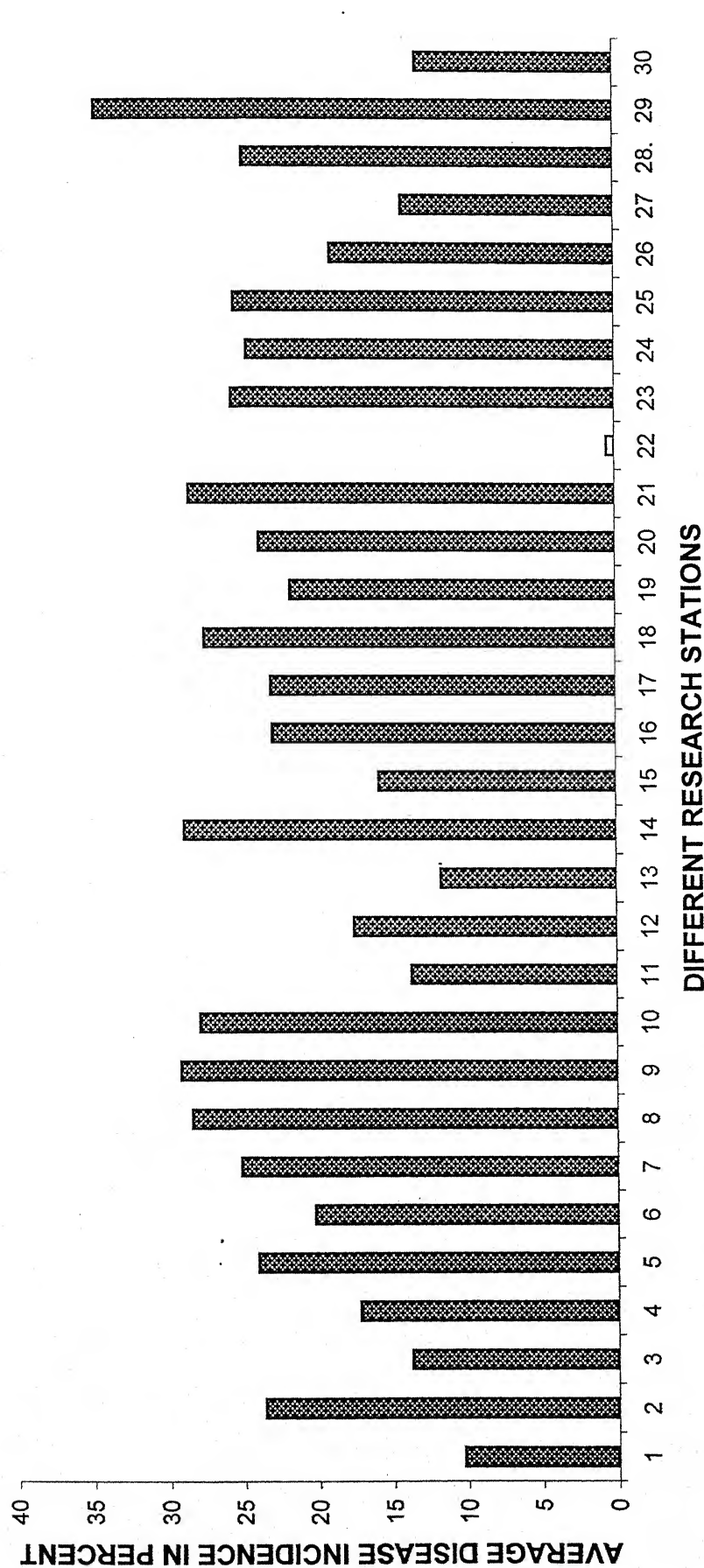
13.	G.P. Pant University Pant Nagar, Nainital	102	12	11.76
14.	Government Agriculture Centre, Atarra, Banda	124	35	29.03
15.	Government Agriculture Centre, Bansi, Banda	88	14	15.90
16.	Government Agriculture Khaptiha, Banda	117	27	23.07
17.	Government Agriculture Research Centre, Bharari, Jhansi	138	32	23.18
18.	Government Agriculture Centre, Bagi, Jalaun	94	26	27.65
19.	Groundnut Research Station, Mainpuri	87	19	21.83
20.	N.D. University Kumarganj, Faizabad	142	34	23.94
21.	Oil Seed Research Farm, Kalyanpur, Kanpur	129	37	28.68
22.	Regional Research Centre, Amroha, Jhansi	—	—	—
23.	Regional Research Station, Uttaripura, Kanpur Dehat	93	24	25.80
24.	Regional Research Centre, Hardoi	117	27	24.78
25.	Regional Research Centre, Varanasi	125	32	25.60
26.	Regional Research Station, Madhurikund, Mathura	89	17	19.10
27.	Regional Research Station, Dilip Nagar, Kanpur	105	15	14.28

FIGURE -5

Prevalence and incidence of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. In the year, 2002 in Kharif crop season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Farm Centre Mauranipur, Jhansi.
11. C.S.Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jaluan.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumarganj, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagar, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE-5



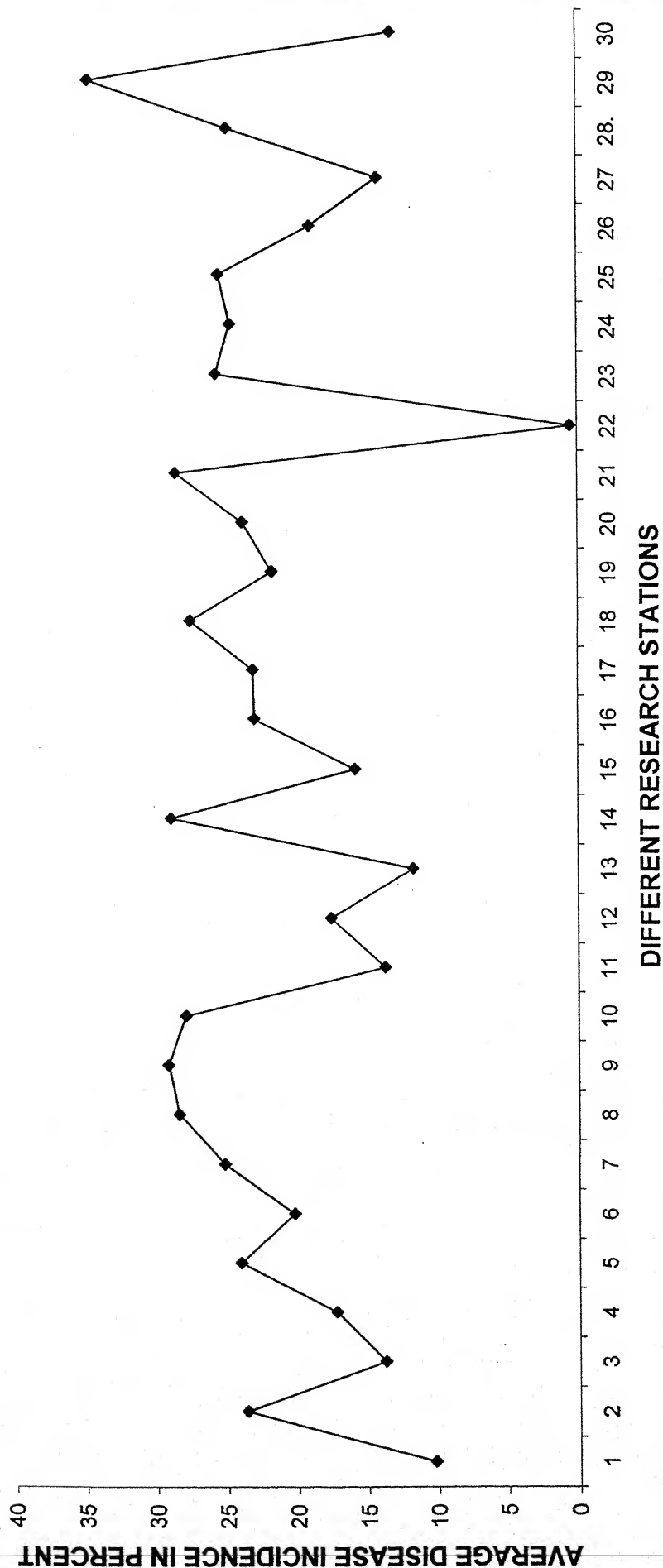
PREVALENCE AND INCIDENCE OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2002 IN KHARIF SEASON.

FIGURE - 6

Prevalence and incidence of Leaf spot Disease of *Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. of the year 2002, in Kharif season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE -6



PREVALENCE AND INCIDENCE OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2002 IN KHARIF SEASON.

28.	Regional Agriculture Demonstration Centre, Meerut	132	33	25.0
29.	Vegetable Research Farm Chandra Shekhar Azad University of Agriculture and Technology, Kanpur	138	47	34.85
30.	Vegetable Research Farm Kalyanpur, Kanpur	173	23	13.29

(-) = Denotes Absence of Disease.

The results presented in Tables IX and X and Figures 3, 4, 5 and 6 revealed that incidence of disease at different research stations during Kharif seasons varied from 13.48 per cent to 31.57 viz., Culture - 7301, Kamgranj Selection - 2, Culture - 9102, Culture - 7015, Culture - 9118, Culture - 7708, H.D-4 Akra Jai, DB-1, Culture - 8403, Todi 125-136, Culture - 7604, Pusa Ealy Prolific, Goyal, JDL - 79, Rajani, Culture - 6801, Culture - 8101, Culture - 7710, Hatikan, HD-66, Culture - 7210, Culture - 9109, Culture - 9113, Culture - 8005, JDL - 85, HD - 93 and Kalyanpur Type 1 and 10.25 per cent to 34.85 per cent from the germplasms/cultures viz., Culture - 7301, Culture - 7210, Akra Jai, Culture - 9118, Culture - 6801, Hatikan, Culture - 8005, Culture - 7015, Tode 125-136, Goya, JDL - 85, Pusa Early Prolific, Culture - 9102, Kamgranj Selection - 2, HD-4, Culture - 9109, Culture - 8101, HD - 66, DPL-1, Culture - 8403, DB-1, HD- 93, Rajani, JDL-79, Culture - 7604, Culture - 7710, Culture - 9913, Culture - 9104 and Kalyanpur Type - 1 in the year, 2001 and 2002 respectively showing its wide spread nature. The maximum disease intensity 31.57 per cent and 34.85 per cent was recorded at Vegetable Research Farm C.S. Azad University of Agriculture and Technology Kanpur in both the years from the germplasm Culture - Kalyanpur Type - 1 followed by 30.80 per cent and 29.26 per cent at Government Agriculture Centre, Jalaun and Crop Research Farm Modipuram Area, Meerut in the years 2001 and 2002 from the

germplasms/cultures HD-93 and Culture - 9113 respectively and rest of the locations, while minimum 13.48 per cent and 10.25 per cent disease incidence was recorded at Agriculture Science Centre Ganiwa, Chitrakoot.

The average disease incidence ranged from 13.48 per cent to 31.57 per cent and 10.25 per cent to 34.85 per cent in both the years of survey at different crop research stations.

In general, it was concluded that maximum disease incidence was recorded at Vegetable Research Farm, C.S. Azad University of Agriculture, Kanpur.

DISEASE INTENSITY -

The intensity of disease, was examined in laboratory by the method described under "Material and Method". The observations recorded are presented in Table XI and XII and Figure 7, 8, 9 and 10.

TABLE - XI

Prevalence and Intensity of Leaf spot Disease of *Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.) Keissler at different Research Stations of U.P. in the year 2001 in Kharif season.

S. No.	Name of Research Station	Location	Germplasm	Disease Intensity in %
1.	Agriculture Science Centre	Ganiwa Chitrakoot	Culture-7301	26.40
2.	Charyar Farm	Bulandshahar	HD-4	31.50
3.	Crop Research Farm	Etawah	Culture-6801	24.90
4.	Regional Research Station	Saini, Allahabad	Culture-7015	34.70
5.	Crop Research Farm	Saraimira, Farrukhabad	Culture-8101	35.30
6.	Crop Research Farm	Baraur, Farrukhabad	JDL-85	35.60

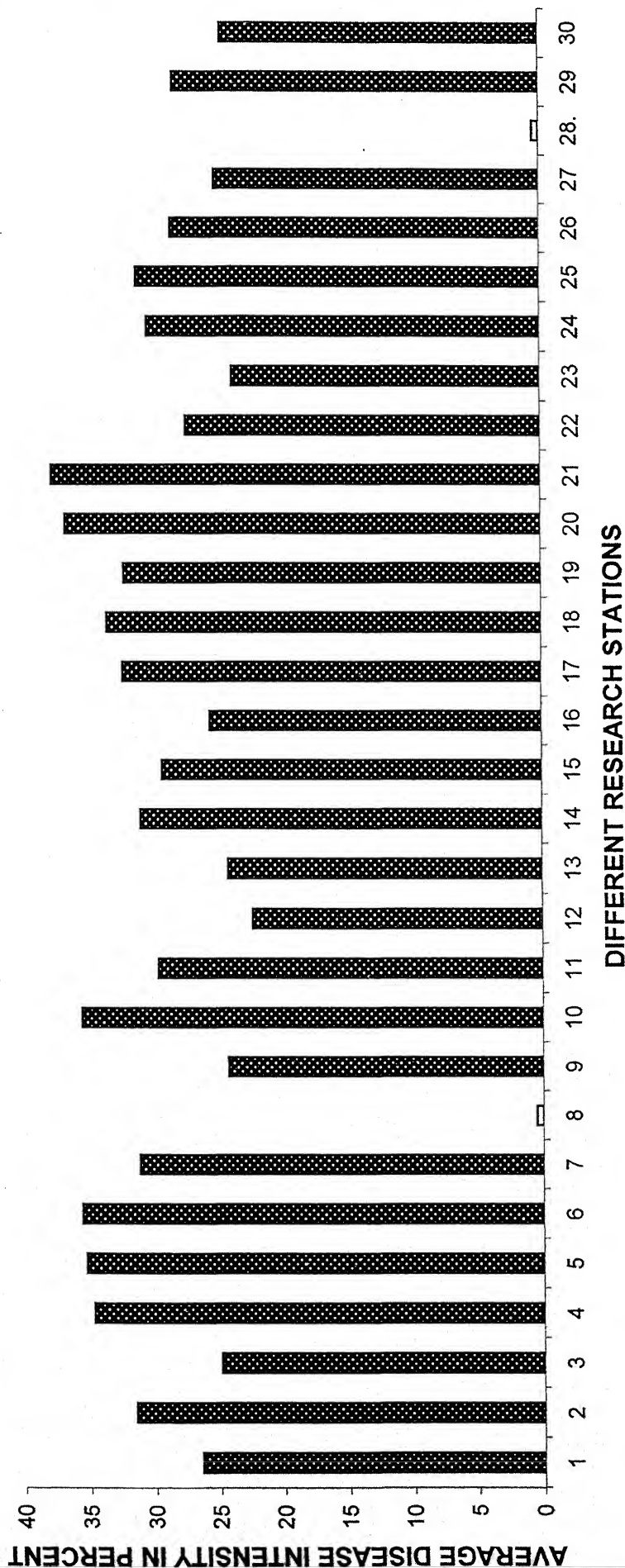
7.	Crop Research Farm	Araul, Kanpur	Culture-8403	31.20
8.	Crop Research Farm	Deegh, Kanpur	Culture-9104	—
9.	Crop Research Farm	Modipuram Area, Meerut	Culture-9113	24.30
10.	Crop Research Centre	Mauranipur Jhansi	Rajani	35.60
11.	C.S. Azad University Research Farm	Belatal, Mahoba	Culture-9118	29.70
12.	Directorate of Vegetable Research Farm	Varanasi	Todi 125-136	22.40
13.	G.P. Pant University Farm	Pant Nagar, Nainital	Culture-7210	24.30
14.	Government Agriculture Centre	Atarra, Banda	Culture-7710	31.10
15.	Government Agriculture Centre	Bansi, Banda	Culture-8005	29.40
16.	Government Agriculture Centre	Khaptiha, Banda	Culture-9102	25.70
17.	Government Agriculture Centre	Bharari, Jhansi	Kamgranj Selection - 2	32.40
18.	Government Agriculture Centre	Bogi, Jalaun	HD-93	33.60
19.	Government Research Station	Mainpuri	Pusa Early Prolific	32.30
20.	N.D. University	Kumarganj, Faizabad	Culture-9109	36.80
21.	Oil Seed Research Farm	Kalyanpur, Kanpur	Culture-7604	37.80
22.	Regional Research Centre	Amroha, Jhansi	Culture-7708	27.50

FIGURE - 7

Prevalence and Intensity of Leaf Spot Disease of *Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. of the year 2001, in Kharif season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE -7



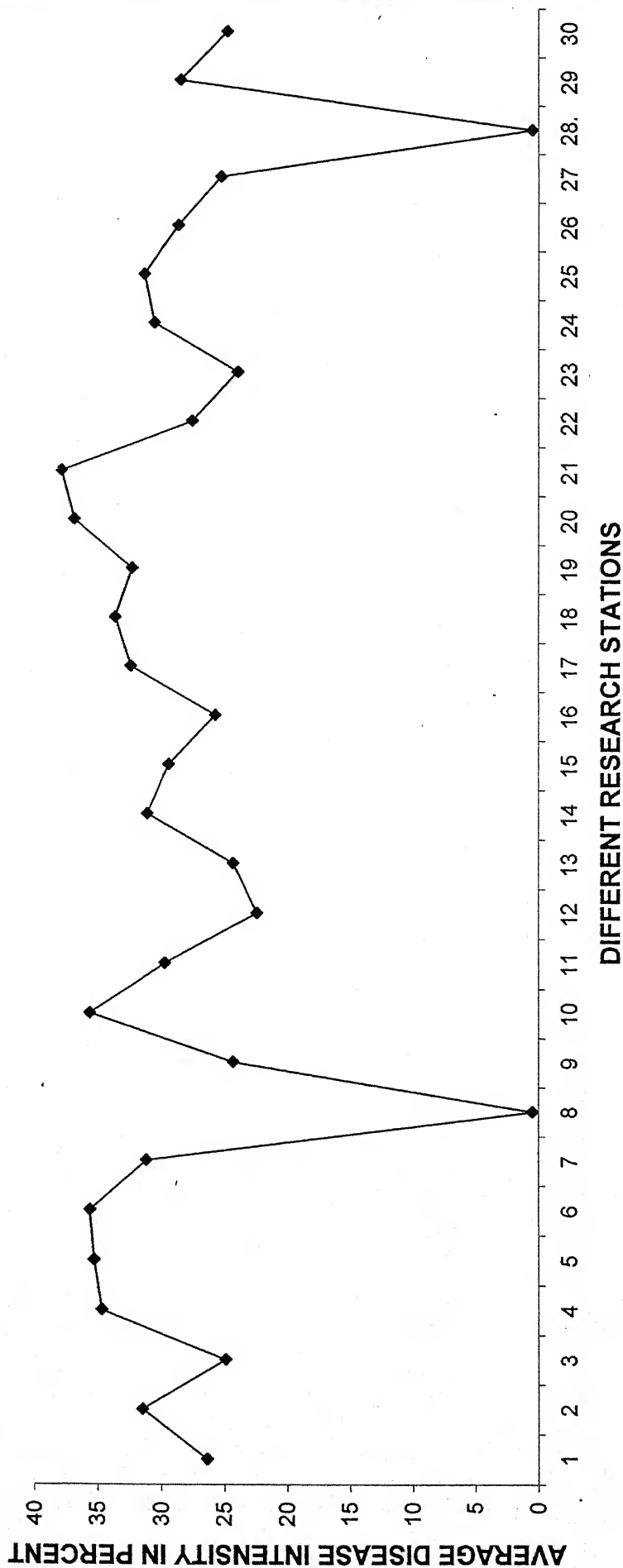
PREVALENCE AND INTENSITY OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2001 IN KHARIF SEASON.

FIGURE - 8

Prevalence and Intensity of Leaf Spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. of the year 2001, in Kharif season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE-8



PREVALENCE AND INTENSITY OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2001 IN KHARIF SEASON.

23.	Regional Research Station	Uttaripura Kanpur Dehat	JDL-79	23.90
24.	Regional Research Station	Hardoi	HD-66	30.50
25.	Regional Research Centre	Varanasi	DB-1	31.30
26.	Regional Research Station	Madhurikund Mathura	Goya	28.60
27.	Regional Research Station	Dilip Nagar Kanpur	Hatikan	25.20
28.	Regional Agriculture Demonstration Centre	Meerut	DPL-1	—
29.	Vegetable Research Farm	C.S. Azad University of Agriculture and Technology Kanpur	Kalyanpur Type - 1	38.40
30.	Vegetable Research Farm	Kalyanpur, Kanpur	Abra Jai	24.70

(—) = Denotes Absence of disease.

TABLE - XII

Prevalence and Intensity of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.) Keissler, at different Research Stations of U.P. in the year 2002 in Kharif season.

S. No.	Name of Research Station	Location	Germplasm	Disease Intensity in %
1.	Agriculture Science Centre	Ganiwa Chitrakoot	Culture-7301	25.30
2.	Charyar Farm	Bulandshahar	HD-4	34.30
3.	Crop Research Farm	Etawah	Culture-6801	30.10


4.	Regional Research Station	Saini, Allahabad	Culture-7015	32.60
5.	Crop Research Farm	Saraimira, Farrukhabad	Culture-8101	39.50
6.	Crop Research Farm	Baraur, Farrukhabad	JDL-85	35.20
7.	Crop Research Farm	Araul, Kanpur	Culture-8403	31.80
8.	Crop Research Farm	Deegh, Kanpur	Culture-9104	23.70
9.	Crop Research Farm	Modipuram Area, Meerut	Culture-9113	37.40
10.	Crop Research Centre	Mauranipur Jhansi	Rajani	31.80
11.	C.S. Azad University Research Farm	Belatal, Mahoba	Culture-9118	20.30
12.	Directorate of Vegetable Research Farm	Varanasi	Todi 125-136	37.40
13.	G.P. Pant University Farm	Pant Nagar, Nainital	Culture-7210	38.20
14.	Government Agriculture Centre	Atarra, Banda	Culture-7710	17.90
15.	Government Agriculture Centre	Bansi, Banda	Culture-8005	28.40
16.	Government Agriculture Centre	Khaptiha, Banda	Culture-9102	20.50
17.	Government Agriculture Centre	Bharari, Jhansi	Kamgranj Selection - 2	28.60
18.	Government Agriculture Centre	Bagi, Jalaun	HD-93	30.40
19.	Government Research Station	Mainpuri	Pusa Early Prolific	31.70


FIGURE - 9

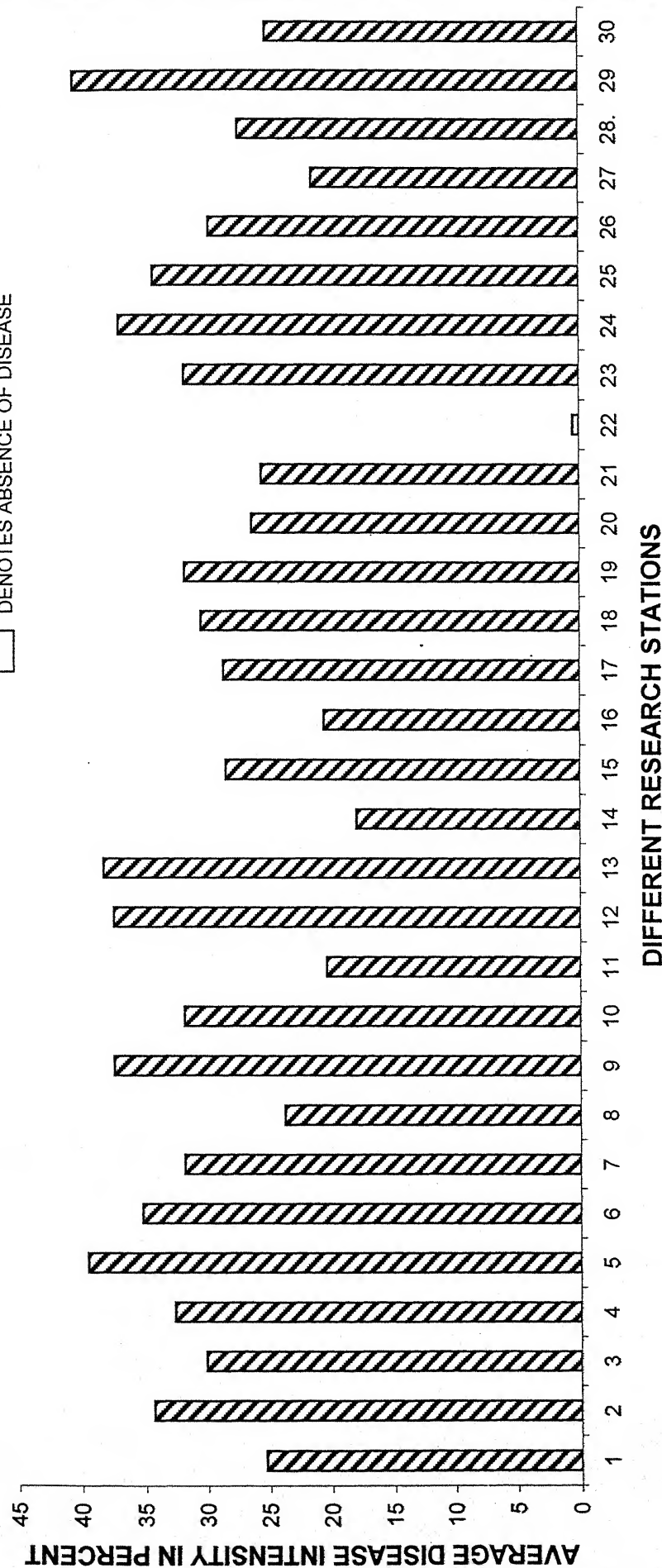
Prevalence and Intensity of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of the year 2001, in Kharif season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut.
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE-9

 DENOTES PREVALENCE AND DISEASE INTENSITY

 DENOTES ABSENCE OF DISEASE



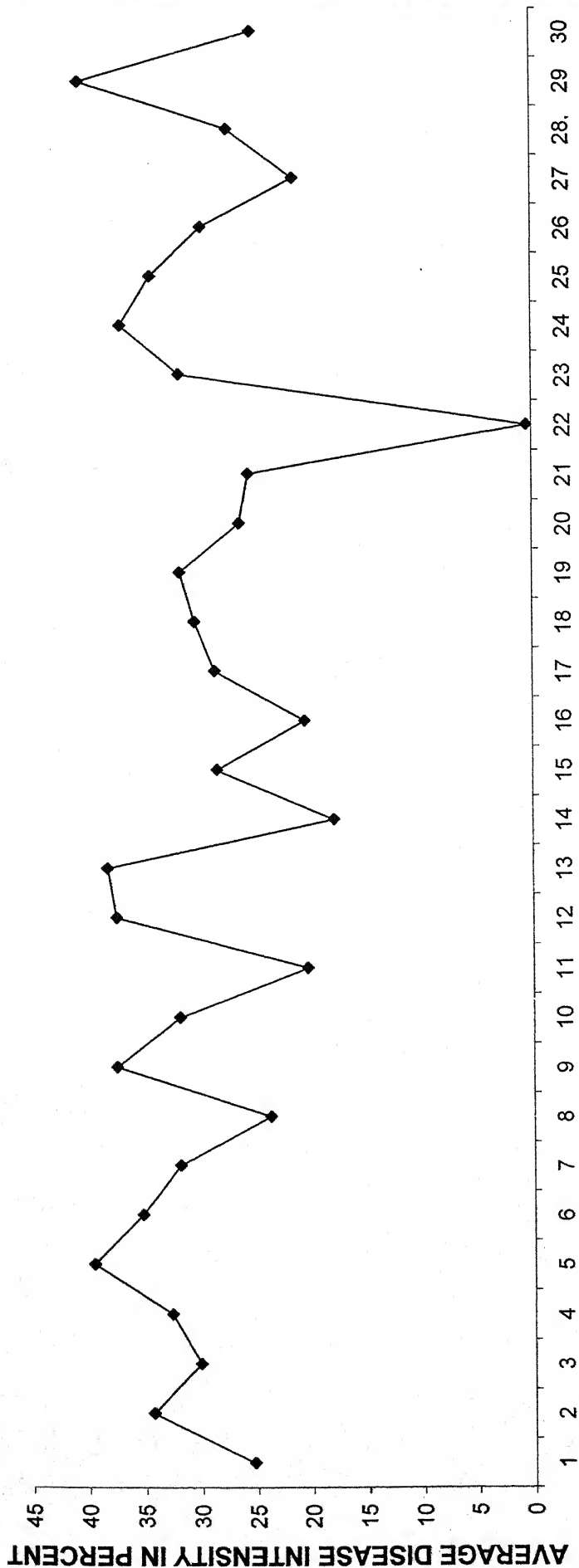
PREVALENCE AND INTENSITY OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2002 IN KHARIF SEASON.

FIGURE - 10

Prevalence and Intensity of Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler at different Research Stations of U.P. of the year 2002, in Kharif season.

1. Agriculture Science Centre, Ganiwa, Chitrakoot.
2. Charayar Farm, Bulandshahar.
3. Crop Research Farm, Etawah.
4. Regional Research Station, Saini, Allahabad.
5. Crop Research Farm Saraimira, Farrukhabad.
6. Crop Research Farm Baraur, Farrukhabad.
7. Crop Research Farm Araul, Kanpur.
8. Crop Research Farm Deegh, Kanpur.
9. Crop Research Farm Modipuram Area, Meerut.
10. Crop Research Centre Mauranipur, Jhansi.
11. C.S. Azad University Research Farm Belatal, Mahoba.
12. Directorate of Vegetable Research Farm, Varanasi.
13. G.P. Pant University Farm Pant Nagar, Nainital.
14. Govt. Agriculture Centre Atarra, Banda.
15. Govt. Agriculture Centre Bansi, Banda.
16. Govt. Agriculture Centre Khaptiha, Banda.
17. Govt. Agriculture Centre Bharari, Jhansi.
18. Govt. Agriculture Centre Bagi, Jalaun.
19. Groundnut Research Station, Mainpuri.
20. N.D. University Kumargang, Faizabad.
21. Oil Seed Research Farm Kalyanpur, Kanpur.
22. Regional Research Centre Amroha, Jhansi.
23. Regional Research Station Uttaripura, Kanpur Dehat.
24. Regional Research Centre, Hardoi.
25. Regional Research Centre, Varanasi.
26. Regional Research Station Madhurikund, Mathura.
27. Regional Research Station Dilip Nagr, Kanpur.
28. Regional Agriculture Demonstration Centre, Meerut
29. Vegetable Research Farm C.S. Azad University of Agriculture and Technology, Kanpur.
30. Vegetable Research Farm Kalyanpur, Kanpur.

FIGURE-10



DIFFERENT RESEARCH STATIONS

PREVALENCE AND INTENSITY OF LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler, AT DIFFERENT RESEARCH STATIONS OF U.P. IN THE YEAR 2002 IN KHARIF SEASON.

20.	N.D. University	Kumarganj, Faizabad	Culture-9109	26.30
21.	Oil Seed Research Farm	Kalyanpur, Kanpur	Culture-7604	25.50
22.	Regional Research Centre	Amroha, Jhansi	Culture-7708	—
23.	Regional Research Station	Uttaripura Kanpur Dehat	JDL-79	31.70
24.	Regional Research Station	Hardoi	HD-66	36.90
25.	Regional Research Centre	Varanasi	DB-1	34.20
26.	Regional Research Station	Madhurikund Mathura	Goya	29.70
27.	Regional Research Station	Dilip Nagar Kanpur	Hatikan	21.40
28.	Regional Agriculture Demonstration Centre	Meerut	DPL-1	27.30
29.	Vegetable Research Farm	C.S. Azad University of Agriculture and Technology Kanpur	Kalyanpur Type - 1	40.50
30.	Vegetable Research Farm	Kalyanpur, Kanpur	Akra Jai	25.10

(—) = Denotes Absence of disease.

The results presented in Table XI and XII and corresponding Figures 7, 8, 9 and 10 revealed that intensity of disease varied from 22.40 per cent to 38.40 per cent from the germplasms/cultures viz; Todi 125-126, JDL-79, Culture - 9113, Culture - 7210, Akra Jai, Culture - 6801, Hatikan, Culture - 9102, Culture - 7301, Culture - 7708, Goya, Culture - 8005, Culture - 9118, HD-66, Culture - 7710, Culture - 8403, DB-1, HD- 4, Pusa Early Prolific, Kamgranj Selection - 2, HD-93, Culture 7015, Culture - 8101, Rajani, JDL-85, Culture - 9109, Culture -

7604 and Kalyanpur Type - 1 and 17.90 to 40.50 per cent from the germplasms/cultures viz; Culture - 7710, Culture - 9118, Culture - 9102, Hatikan, Culture - 9104, Akra Jai, Culture - 7301, Culture - 7604, Culture - 9109, DPL-1, Culture - 8005, Kamrganj, Selection - 2, Goya, Culture - 6801, HD-93, Pusa Early Prolific, JDL-79, Rajani, Culture - 8403, Culture - 7015, DB-1, HD-4, JDL-85, HD- 66, Culture - 9113, Todi 125-136, Culture - 7210, Culture - 8101 and Kalyanpur Type - 1 respectively, showing wide spread prevalence in nature in the years 2001 and 2002. Maximum 38.40 per cent and 40.50 per cent disease intensity was recorded from the germplasm/culture, "Kalyanpur Type - 1" at Chandra Shekhar Azad University of Agriculture and Technology, Kanpur, followed by 37.80 per cent and 39.50 per cent from Oil Seed Research Farm Kalyanpur, Kanpur and Crop Research Farm Saraimira, Farrukhabad from germplasms/cultures 7604 and 8101 and rest of locations in both the years 2001 and 2002, while minimum 22.40 per cent and 17.90 per cent disease intensity was recorded from Directorate of Vegetable Research Farm, Varanasi and Government Agriculture Centre, Atarra, Banda from the germplasms/cultures Todi 125-136 and Culture - 7710 in both the years respectively.

In general, it was concluded that maximum disease intensity was recorded from Chandra Shekhar Azad University of Agriculture and Technology, Kanpur.

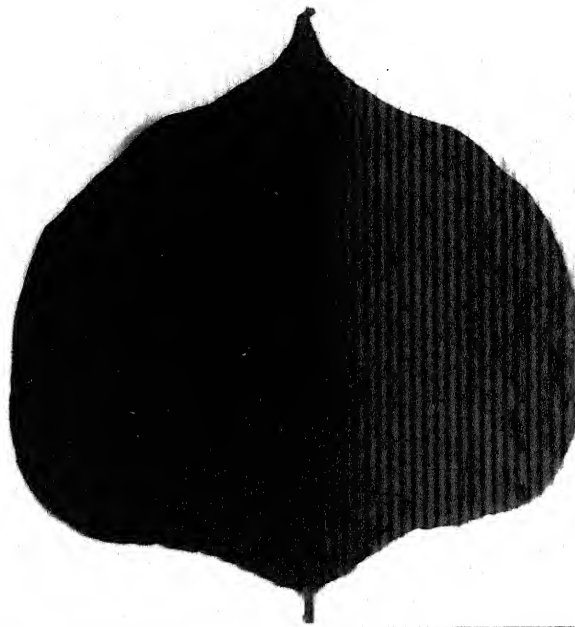
The disease incidence as well as disease intensity could not be recorded in the years 2001 and 2002 at Crop Research Farm Deegh, Kanpur and Regional Agriculture Research Station Dilip Nagar, Kanpur and Regional Research Centre, Amroha, Jhansi respectively.

SYMPTOMATOLOGY OF THE DISEASE -

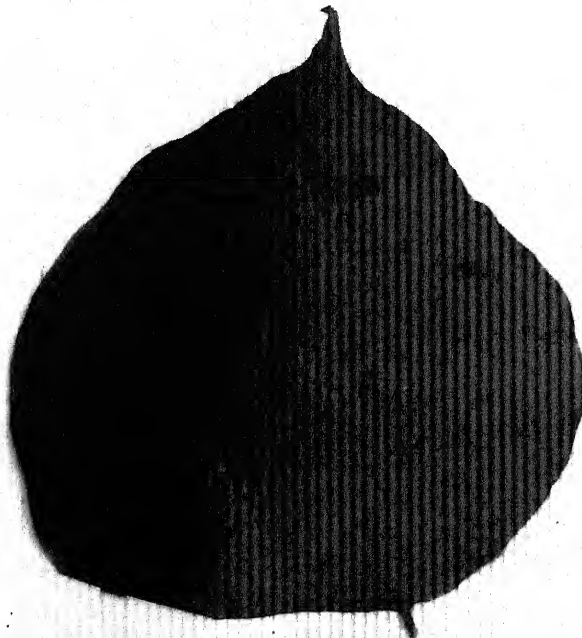
The disease was found prevalent in the month of December and January in Rabi sown crop, when the plants attained the age of two to three months.

The disease appeared in the form of numerous small spots on the upper surface of leaves. A few leaves were found studded all over these spots. The

PLATE -II
(a)



(b)



Symptoms of the disease leaf spot of Dolichos bean (*Dolichos lablab*, L.)
on different leaves caused by *Alternaria alternata* (Fries.), Keissler

spots were recorded to black; circular to oval with paler margins and yellow hallow measuring 0.15 - 1.0 cm. in size; with characteristic concentric rings and cracked centre. The spots at first, were recorded as smaller in size. In case of serious attack the spots were found numerous and extending along the whole surface due to coalescence of adjacent pustules. In late stage of the disease due to severe infection the affected spots become perforated irregularly due to falling away of dead tissue causing short holes. In general the leaves of matured plants exhibited the disease symptoms. The symptoms of the disease caused by *Alternaria alternata* (Fries), Keissler, are gives in Plate No. II.

ISOLATION AND PURIFICATION OF THE PATHOGEN -

The isolations were made fom the leaf spots of Dolichos bean varieties collected from different localities, showing characteristic symptoms of the disease, causing Alternaia leaf spot by the technique described under "Material and Method". The pathogen was isolated on 2.0 per cent Potato Dextrose Agar medium by transferring sterilized bits of diseased leaf tissue. The isolates were further purified by single spore culture technique. All the isolates were found morphologically similar and pure cultures of the pathogen, thus obtained were maintained on Potato dextrose Agar medium and utilized for different studies under investigation.

PATHOGENICITY TEST -

The pathogenicity of purified isolate of *Alternaria alternata* (Fries.) Keissler was tested on 45 days old seedling of high susceptible germplasm/culture according to the method described under "Material and Method". The plants were raised from the surface sterilized seeds in pots filled wth sterilized soil and were inoculated with spore-cum-mycelial suspension of the fungus on injured and un-injured leaves of Dolichos bean. The inoculated potted plants were kept in humid chambers for 48 hours and thereafter taken out. The observations regarding development of disease and symptoms due to pathogenic attack, were recorded as summarised in Table XIII and Figures 11 and 12.

TABLE-XIII

Pathogenicity of *Alternaria alternata* (Fries.), Keissler, on leaves of Dolichos bean (*Dolichos lablab*, L.).

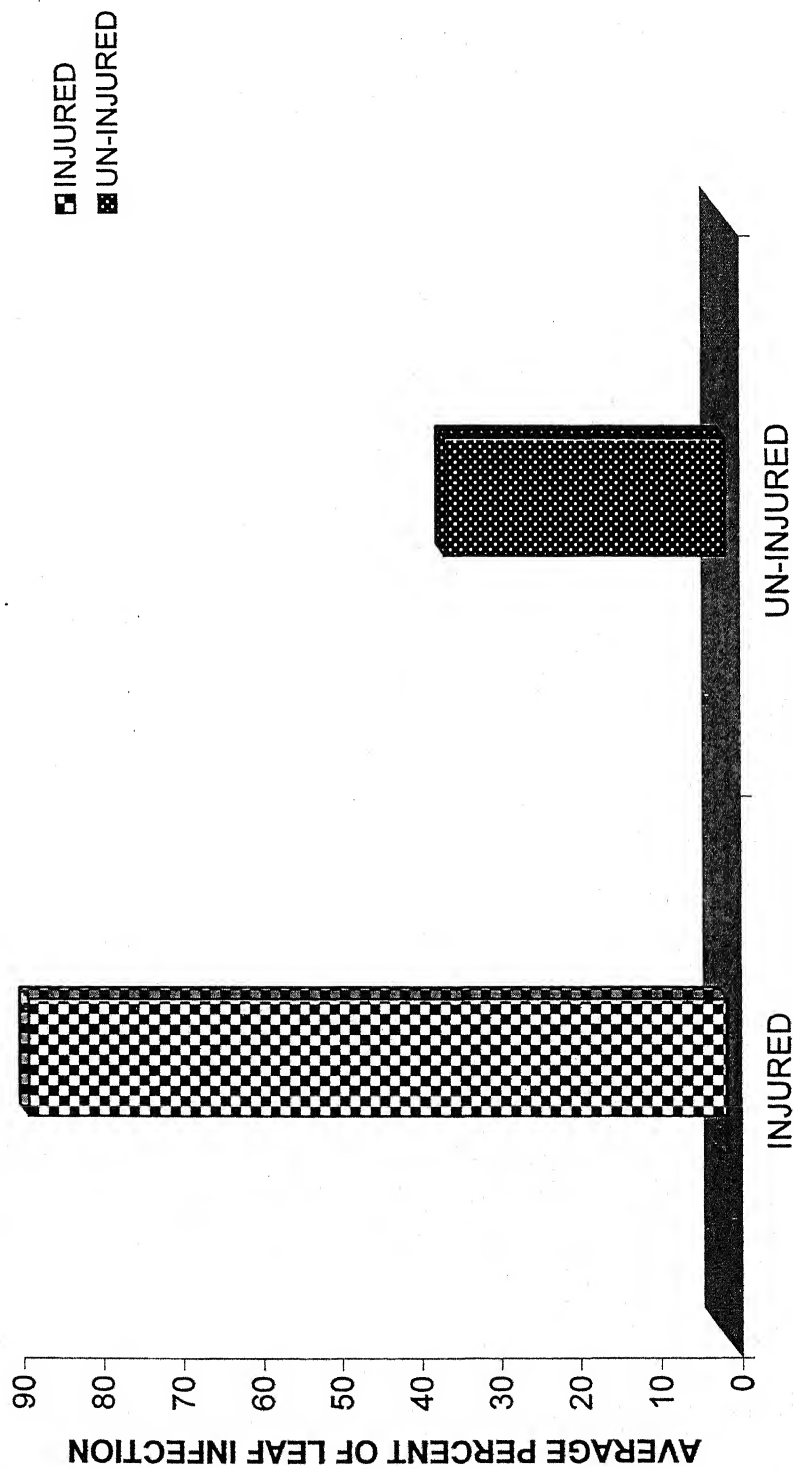
Mode of Inoculation	Treat-ments		Number of Leaves inoculated		Number of Leaves showing		Leaves Infection in %		Average percentage of Infection	
	Inju-red	un-inju-red	Inju-red	un-inju-red	Inju-red	un-inju-red	Inju-red	un-inju-red	Inju-red	un-inju-red
Inoculated with fungal suspension	Pot-1	Pot-1	10	10	8	3	80	30	87.50	35.0
	Pot-2	Pot-2	10	10	9	4	90	40		+
	Pot-3	Pot-3	10	10	10	2	100	20		
	Pot-4	Pot-4	10	10	8	5	80	50		
Control	Pot-1	Pot-1	10	10	00	00	00	00		
	Pot-2	Pot-2	10	10	00	00	00	00	00	00

The results given in Table XIII and corresponding Figures 11 and 12 indicated that the pathogen, *Alternaria alternata* (Fries.), Keissler, was capable to infect both injured and un- injured leaves. The infection percentage of 87.50, was recorded on injured leaves in comparison to un-injured leaves, which exhibited 35.0 infection percentage and proved the fact that injured leaves provided avenues for the pathogenic attack. The infection percentage was higher in the injured leaves in comparison to un-injured leaves.

The initial disease symptoms appeared after 3-4 days of inoculation in the form of small; scattered; brown; circular to irregular spots, which gradually became large in size measuring 2.0 mm. to 5.0 mm. in diameter after six days of inoculation. Later these spots became coalesced to form bigger spots giving the leaves a blighted appearance. The symptoms recorded, were similar as recorded under natural conditions. The leaves sprayed with distilled water served as control and remained free from disease.

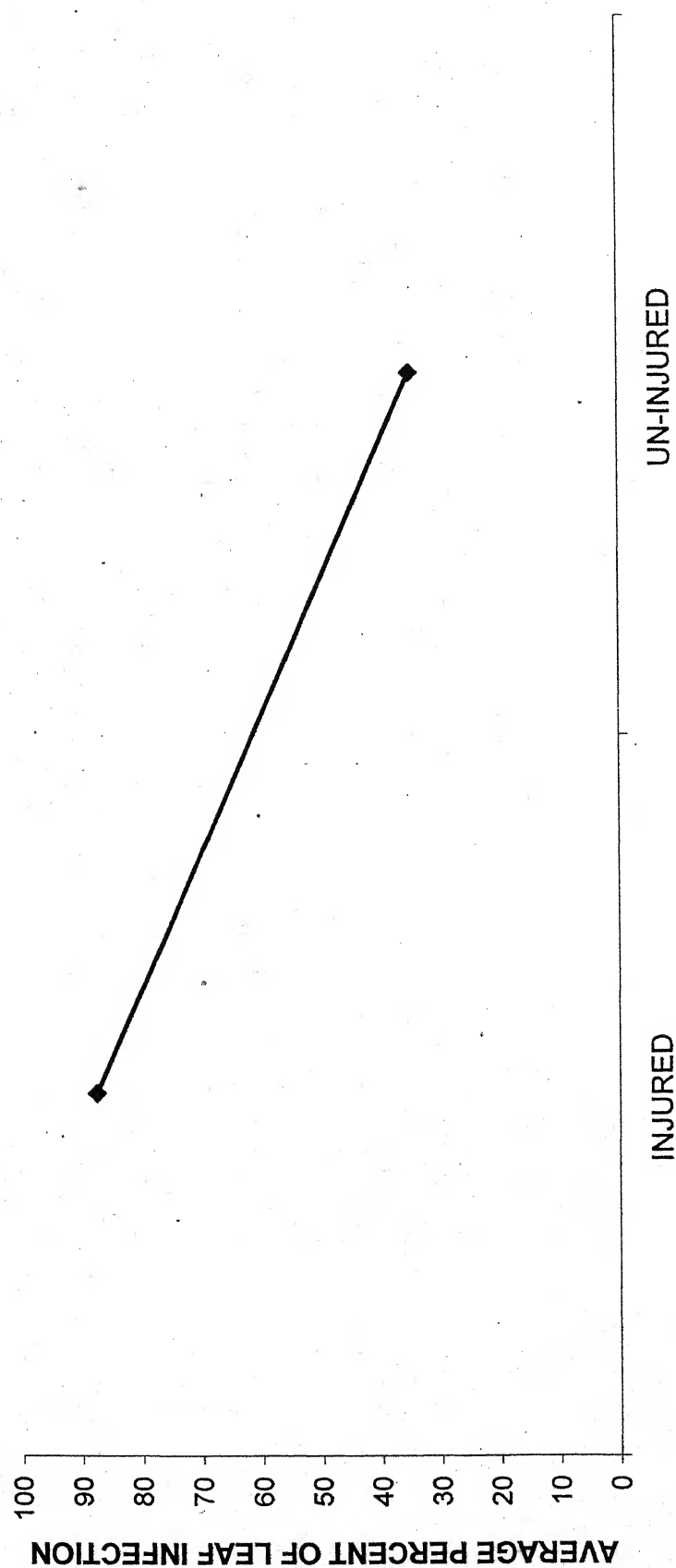
Reisolation from artificially leaf spots produced the same fungus,

FIGURE -11



PATHOGENICITY OF *Alternaria alternata* (Fries.), Keissler, ON LEAVES OF *DOLICHOS BEAN*
(*Dolichos lablab*, L.)

FIGURE-12



PATHOGENICITY OF *Alternaria alternata* (Fries.), Keissler, ON LEAVES OF DOLICHOS BEAN
(*Dolichos lablab*, L.)

Alternaria alternata (Fries.), Keissler, which was previously isolated from the naturally infected Dolichos bean leaves and used for inoculation. The isolations, inoculations, pathogenicity and reisolations proved the Koch's postulates.

MORPHOLOGICAL CHARACTERS AND IDENTIFICATION OF THE PATHOGEN, *Alternaria alternata* (Fries.), Keissler.

Morphological characters are considered to be the main criterion for identifying the species in a particular group of fungi. With this aim, the pathogen under study was identified on the basis of morphological characters on Potato dextrose agar medium as described below according to the technique described in "Material and Method" and given in Plates No. III and IV.

COLONY -

Colonies usually found moderately fast growing, which in the beginning, were dull white; fluffy; circular and later turned into dark greenish olive with abundant sporulation.

MYCELIUM -

Mycelium was found as aseptate and branched. In earlier stages it was hyaline, later turning into black and olive buff in colour measuring 3.20 - 8.60 μ in width.

CONIDIOPHORES -

Conidiophores arise singly or in groups. They were usually simple; septate; straight or bent; sometimes branched; swollen terminally; geniculate and dark olive buff measuring 24.50 - 68.30 \times 3.20 - 6.50 μ in size.

CONIDIA -

Conidia were found formed in chains of 3-15. They were muriform; ovoid to obclavate; obpyriform; dark olive buff in colour and smooth, sometimes verrucose with age. Conidia are provided with 1-5 transverse septa and 0-6 longitudinal septa; measuring 16.70 - 40.50 \times 8.10 - 12.40 μ . The beaks were short; light olive buff in colour and conical or cylindrical measuring 4.70 - 16.90

× 3.20 - 5.70 μ in size with 0-2 transverse septa.

CHLAMYDOSPORES -

The chlamydospores, were found terminal; intercalary and dark olive buff in colours measuring 11.70 - 20.90 μ in diameter.

On the basis of morphological characters the fungus under study is identified as *Alternaria alternata* (Fries.), Keissler, (Keissler, 1912), Syn. *Alternaria tenuis* (Nees.) and also with that Bose (1942) for *Alternaria tenuis* causing Alternaria leaf spot of Sunflower (*Helianthus annuus*, L.) According to Neergaard (1945), Simmons (1967) and Ellis (1971), the fungus was also similar.

EFFECT OF THE MEDIA ON GROWTH AND SPORULATION OF THE PATHOGEN -

Growth of Pathogen on Solid Media -

The fungus was grown on non-synthetic semi-synthetic and synthetic media and observations were noted after incubation of 10 days at 25±1°C. The average diameter of fungal colonies on different media and extent of sporulation are summarised in Table XIV and Figures 13 and 14.

TABLE XIV

Radial growth and sporulation of *Alternaria alternata* (Fries.), Keissler, on different solid media at 25±1°C after 10 days of incubation.

S. No.	Media (Solid)	Average Diameter of colony (mm.)	Sporulation
1.	Asthana and Hawker's Agar	28.50	+
2.	Brown's Agar	17.30	—
3.	Coon's Agar	82.40	+++
4.	Corn Meal Agar	41.20	+
5.	Czapek's Agar	85.60	++++
6.	Leaf Decoction Agar	68.20	+++

7.	Malt Salt Agar	57.30	++
8.	Potato Dextrose Agar	92.40	++++
9.	Richard's Agar	86.70	++++
10.	Sabouraud's Agar	53.10	++

- (-) = Denotes absence of growth.
 (+) = Denotes poor growth.
 (++) = Denotes fair growth.
 (+++) = Denotes good growth.
 (++++) = Denotes excellent growth.

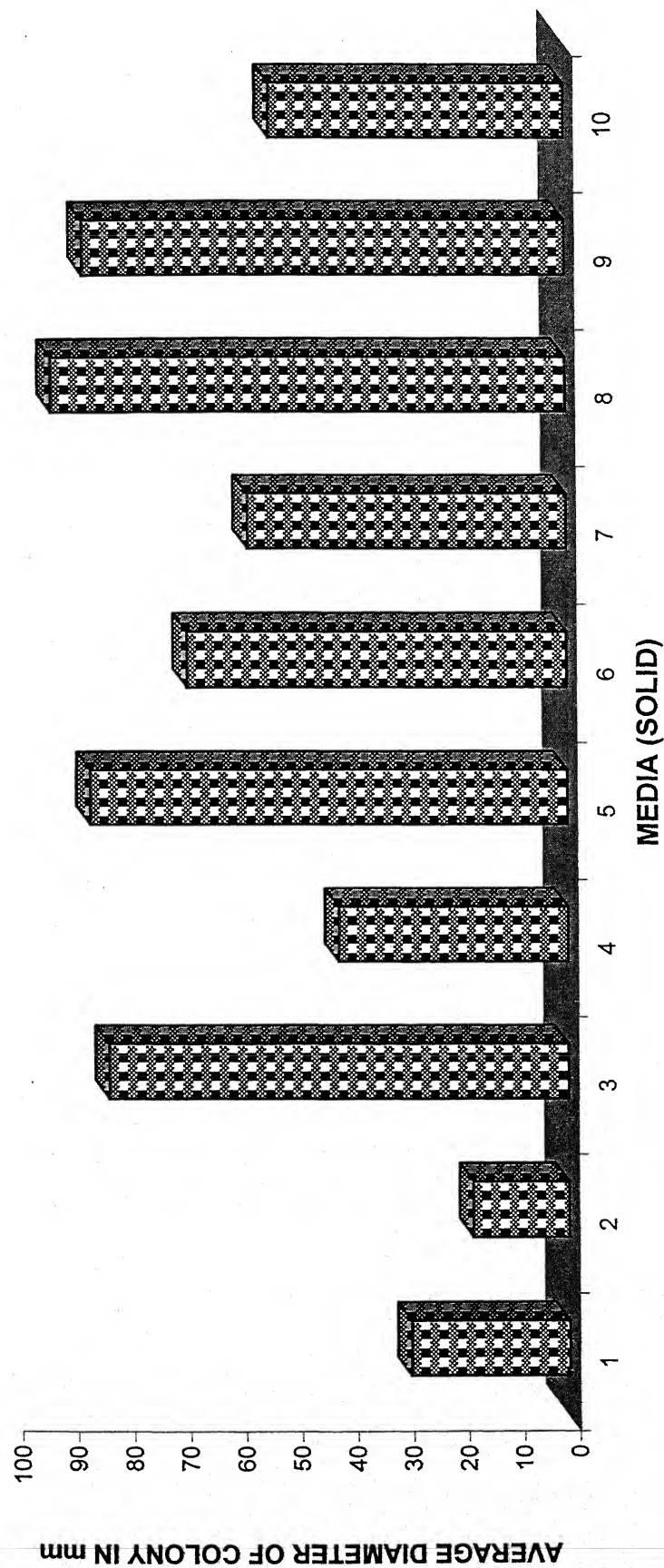
It is evident from the Table XIV given and corresponding Figures 13 and 14, that the best growth of the fungus measuring 92.40 mm. was observed on Potato dextrose agar medium followed by Richard's agar, Czapek's agar, Coon's agar, leaf decoction agar, Malt salt agar, Sabouraud's agar, Corn meal agar and Asthana and Hawker's agar media. The least minimum growth measuring 17.40 mm. was observed on Brown's agar medium.

Potato dextrose agar medium induced excellent sporulation of the fungus. The next were Czapek's agar and Richard's agar media, which also supported excellent sporulation. The sporulation was found good on Coon's agar and Leaf decoction agar medium; fair on Corn Meal agar; Malt salt agar and Sabouraud's agar media, while poor on Asthana and Hawker's agar medium. The pathogen proved as failed, as was unable to sporulate on Brown's agar medium.

CULTURAL CHARACTERS OF THE PATHOGEN :

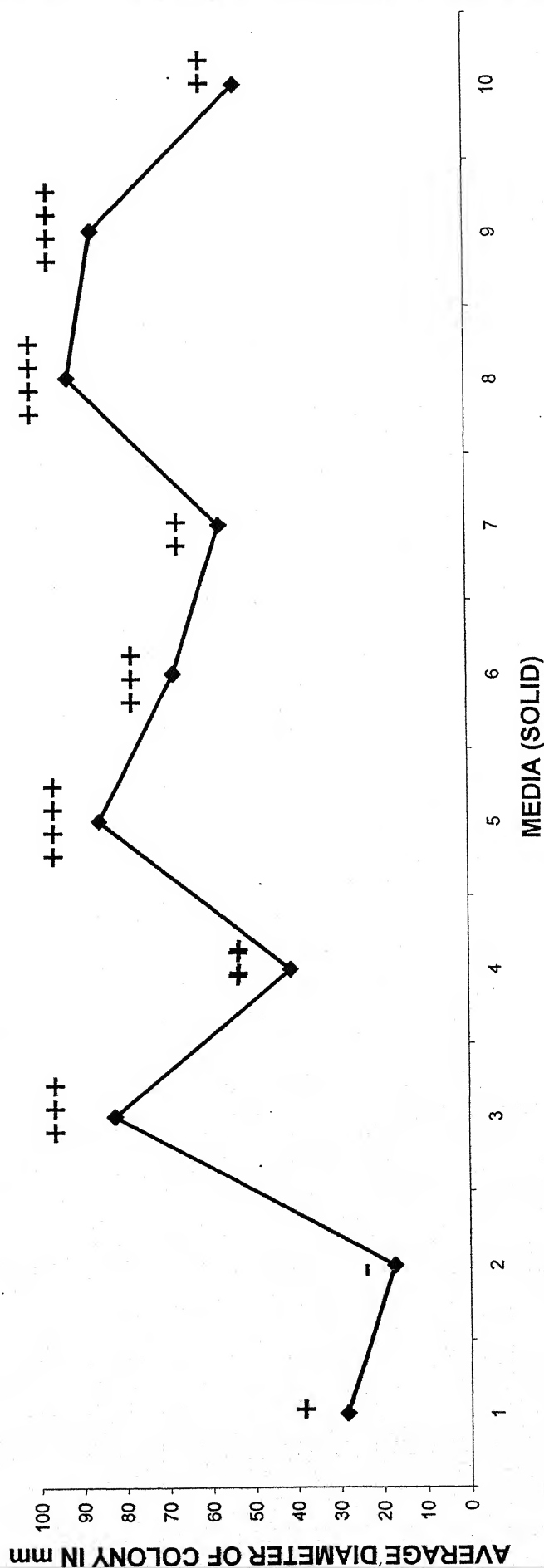
Apart from studying the radial growth and sporulation of the pathogen various other cultural characters viz., growth; shape; zonation and colour of colony; substratum colour; pigmentation; colour of hyphae; colour of conidiophores; number and septation of conidia; variation in shape; size and colour existence and size of chlamydospores produced by the pathogen of different solid media were recorded as given in Table XV (a, b and c) and Plates No. III and IV.

FIGURE-13



RADIAL GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler, ON DIFFERENT SOLID MEDIA AT 25±1°C AFTER 10 DAYS OF INCUBATION.

FIGURE-14



- = DENOTES ABSENCE OF GROWTH
 + = DENOTES POOR GROWTH
 ++ = DENOTES FAIR GROWTH
 +++ = DENOTES GOOD GROWTH
 ++++ = DENOTES EXCELLENT GROWTH

RADIAL GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler, ON DIFFERENT SOLID MEDIA AT $25 \pm 1^\circ\text{C}$ AFTER 10 DAYS OF INCUBATION.

The results concluded in Table XVII (a, b and c) as well as Plates No. 3 and 4 will reveal that colony characters of the pathogen are quite distinct on different types of media under study. The colony growth was found good and compact on Potato dextrose agar medium; good; compact and raised on Czapek's agar medium; good and compact with downy appearance on Richard's agar medium; good sparse; thin and cottony on leaf decoction agar medium; good sparse with entire margin on Coon's agar medium; average sparse with suppressed hairy margin on Corn Meal agar medium; good and semi-suppressed on Malt Salt agar medium; average compact, thin and cottony on Sabouraud's agar medium; average sparse with entire margin on Asthana and Hawker's agar medium and poor sparse with entire margin growth on Brown's agar medium.

The colony shape was recorded almost circular on all the medium except Czapek's agar medium, which exhibited lobe shaped colony.

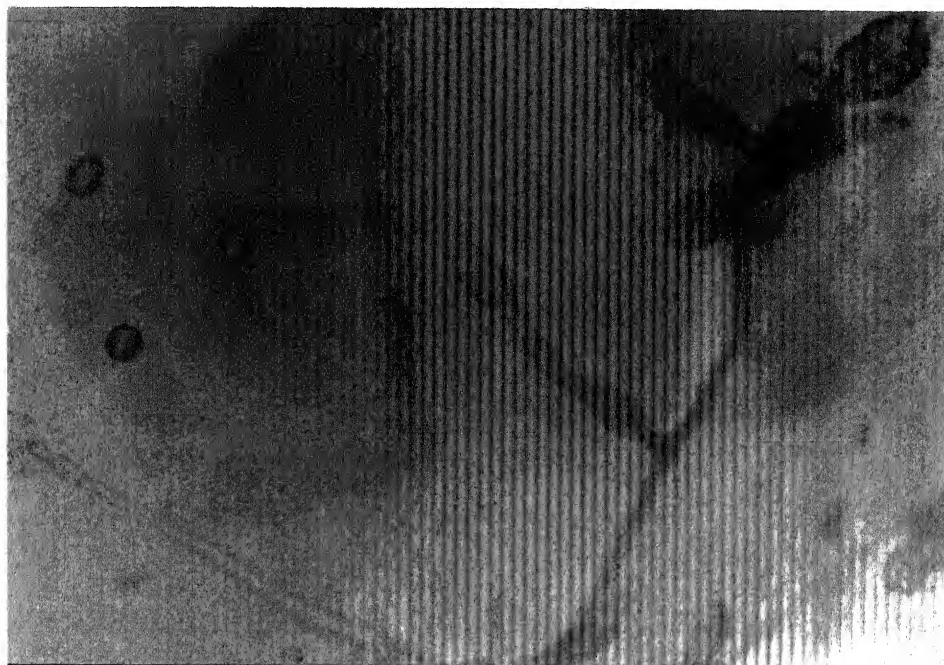
The colour of colony was almost dark grey on Asthana and Hawker's agar medium; green with greenish tinge at the marginal ends on Brown's agar medium; smoky grey on Sabouraud's agar medium; dark black on Richard's agar medium; dark greenish with darker center on Potato dextrose agar medium; dark greenish with whitish margin on leaf decoction agar medium; light green on Corn Meal agar medium and creamy white on Czapek's agar medium and Coon's agar medium.

The substratum colour was also found variable on different types of media viz; iron grey on Potato dextrose agar medium; grey on Brown's agar medium; olive grey on Asthana and Hawker's agar medium; light green on Corn Meal agar medium; blackish green grey on leaf decoction and Malt salt agar media; light vinaceous cinnamon on Richard's agar medium; dark quaker drab on Sabouraud's agar medium and white smoky grey on Coon's agar medium. Zonation was found as distinct on Potato dextrose agar and Richard's agar media; not clear on Asthana and Hawker's agar medium; clear from upper side on Coon's agar and Czapek's agar media; clear from upper side on Coon's Meal

PLATE -III

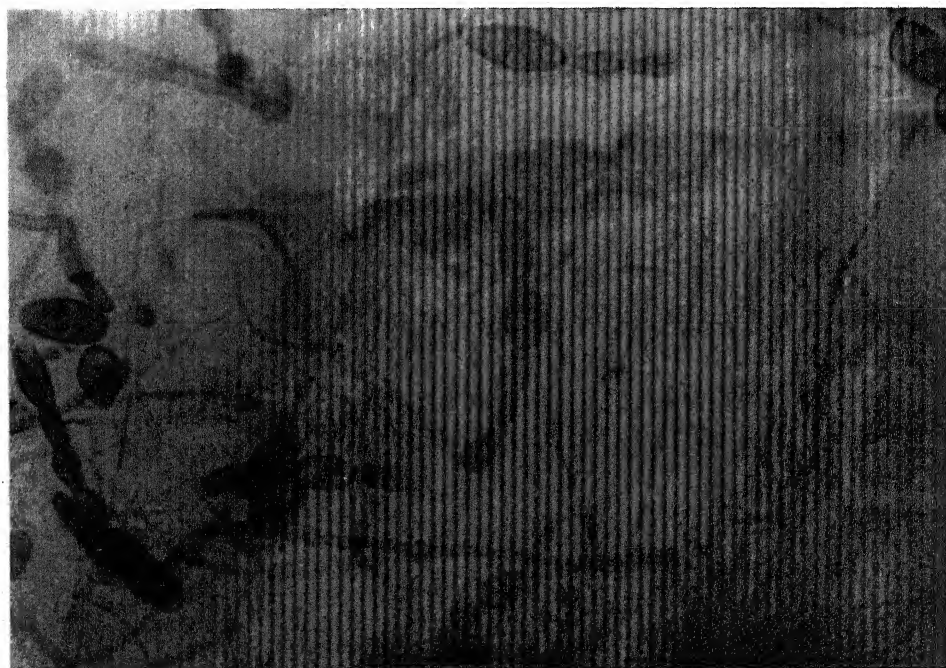
Conidial character of *Alternaria alternata* (Fries.), Keissler

(a)



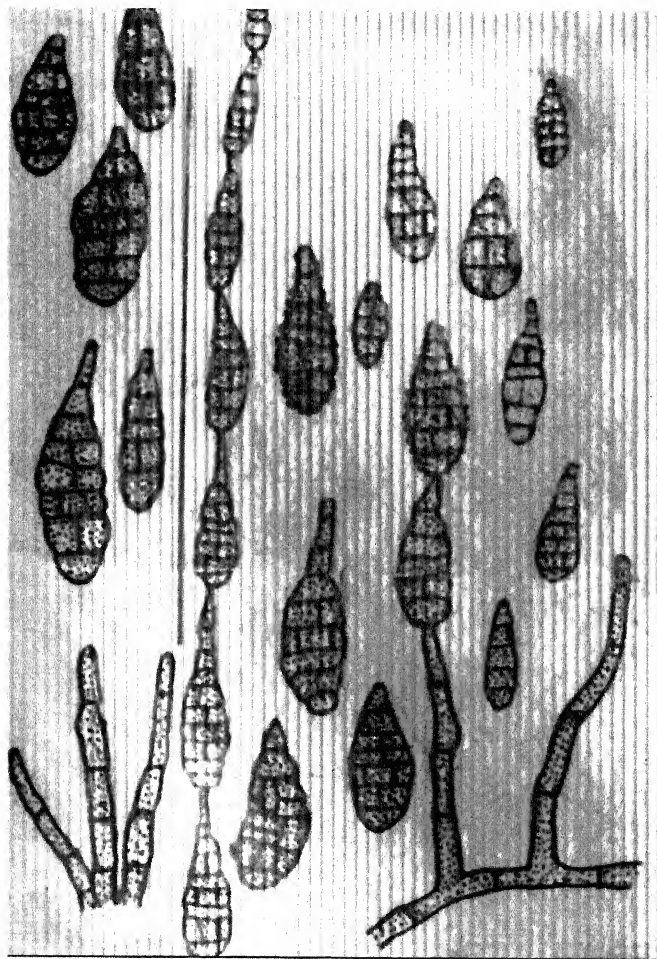
Conidia in chains

(b)



Conidia in chains

PLATE -IV



Conidia of *Alternaria alternata* (Fries.), Keissler in chains

agar, leaf decoction agar and Malt salt agar media; less clear from bottom side and absent from upper side on Brown's agar medium and clear from underside on Sabouraud's agar medium, while pigmentation was found absent on all the different types of media.

Colour of hyphae was recorded as olive buff on Czapek's agar, Richard's agar and Sabouraud's agar media; mid olive on Brown's agar medium; olive buff to greyish on Malt salt agar medium; pale olive buff on Asthana and Hawker's agar medium; pale olive grey on Coon's agar medium; light olive grey on Corn Meal agar and leaf decoction agar media and colourless to greyish on Potato dextrose agar medium. Hyphae were found septate and varied in size from 3.0 - 8.0 μ in width on different types of media under study.

Conidia were also found variable in colours viz; olivaceous to dark brown on Czapek's agar, Malt salt agar, Richard's agar and Sabouraud's agar media; olivaceous to brown on Potato dextrose agar medium; dark olive brown on Asthana and Hawker's agar medium; Olive buff to brown on Brown's agar medium; light brown on Coon's agar medium; dark olive grey on Corn meal agar medium and deep olive brown on leaf decoction agar medium.

Colour of conidiophores was recorded as olive buff to brown on Brown's agar and Richard's agar media; mid olive to brown on Czapek's agar and Sabouraud's agar media; olivaceous brown on Coon's agar and Malt salt agar media; dark olive buff on Asthana and Hawker's agar medium; Pale to olive brown on Potato dextrose agar medium; dark olive brown on leaf decoction agar medium and olive grey on Corn meal agar medium. The conidiophores varied in size from 24.20 - 68.60 \times 3.10 - 7.8 μ on different types of media under study.

Number of conidia borne on conidiophores in chains were also found variable on different types of media viz., 2-5 on Asthana and Hawker's agar; Brown's agar and Coon's agar media; 2-6 on Corn Meal agar; Czapek's agar and Sabouraud's agar media; leaf decoction agar and Malt salt agar media; 2-7 on Richard's agar medium and 3-7 on Potato dextrose agar medium.

TABLE - XV (a)

Comparative study of cultural characters (colony growth, shape, colour, zonation, pigmentation and colour of substrate) of pathogen *Alternaria alternata* (Fries.), Keissler, causing leaf spot disease of Dolichos bean (*Dolichos lablab*, L.) on different types of media.

S. No.	Media	Colony Growth	Colony Shape	Colony Colour	Substratum Colour	Zonation	Pigment
1.	Asthana and Hawker's Agar	Average sparse with entire margin	Circular	Dark grey	Olive grey	Not clear	Absent
2.	Brown's Agar	Poor, sparse with entire margin	Circular	Green with greenish tinge at the marginal ends.	Grey	Less clear from bottom side and absent from upper side	Absent
3.	Coon's Agar	Good, sparse with entire margin	Circular	Creamy white	White smoke grey	Clear from upperside	Absent
4.	Corn Meal Agar	Average, sparse with suppressed hairy margin	Circular	Light green	Light green	Clear from underside	Absent
5.	Czapek's Agar	Good, compact and raised	Lobed	Creamy white	Blackish green grey	Clear from upperside	Absent
6.	Leaf Decoction Agar	Good, sparse, thick and cottony	Circular	Dark green with whitish margin	Olivaceous black	Clear from underside	Absent

7.	Malt Salt Agar	Good and semi-suppressed	Circular	Dark, Black with concentric rings of alternate light green and green colour	Olivaceous Black	Clear from underside	Absent
8.	Potato Dextrose Agar	Good and compact	Circular	Dark greenish darker at the centre	Iron grey	Distinct	Absent
9.	Richard's Agar	Good, compact and downy appearance	Circular	Dark black	Light vinaceous cinnamon	Distinct	Absent
10.	Sabouraud's Agar	Average, compact, thin and cottony	Circular	Smoky grey	Dark quaker drab	Clear from underside	Absent

TABLE - XV (b)

Comparative study of cultural characters (size and colour of hyphae, intercalary or terminal chlamydospores size and colour of conidiophores) of pathogen, *Alternaria alternata* (Fries.), Keissler, causing leaf spot disease of Dolichos bean (*Dolichos lablab*, L.) on different types of media.

S. No.	Media	Hyphae			Chlamydospores			Conidiophore	
		Size	Colour	Septation	Colour	Intercalary of terminal	Size	Size	Colour
1.	Asthana and Hawker's Agar	3.50-5.20 μ	Pale olive buff	Septate	Dark olive green	Terminal and intercalary	5.70 to 11.50 μ	27.0-42.60 \times 4.1-5.40 μ	Dark olive buff
2.	Brown's Agar	3.80-6.40 μ	Mid olive	Septate	Olive Buff to brown	Terminal	86.80 to 12.90 μ	27.50-40.90 \times 4.50-5.10 μ	Olive Buff to Brown
3.	Coon's Agar	3.0-5.60 μ	Pale olive grey	Septate	Light brown	Terminal and intercalary	10.30-15.50 μ	22.20-41.60 \times 4.0-5.40 μ	Olivaceous brown
4.	Corn Meal Agar	3.30-5.70 μ	Light olive grey	Septate	Olive grey	Terminal and intercalary	5.20-12.20 μ	26.0-43.60 \times 3.10-5.30 μ	Olive grey
5.	Czapek's Agar	3.50-7.30 μ	Olive buff	Septate	Dark brown	Terminal and intercalary	10.40-18.50 μ	20.20-61.80 \times 4.60-6.40 μ	Mid olive to brown
6.	Leaf Decoction Agar	3.30-6.10 μ	Light olive grey	Septate	Dark olive brown	Intercalary	9.0-13.50 μ	25.80-48.20 \times 3.60-5.90 μ	Dark olive brown
7.	Malt Salt Agar	3.60-6.40 μ	Olive buff to greyish	Septate	Olivaceous to dark brown	Terminal and intercalary	12.0-20.20 μ	25.80-49.50 \times 4.54-7.63 μ	Olivaceous brown
8.	Potato Dextrose Agar	3.70-8.0 μ	Colourless to greyish	Septate	Olive buff	Terminal and intercalary	13.70-22.80 μ	28.80-68.60 \times 5.70-7.80 μ	Pale to olive brown
9.	Richard's Agar	3.0-7.20 μ	Olive Buff	Septate	Light brown	Terminal and intercalary	12.20-21.30 μ	23.60-66.50 \times 4.80-7.50 μ	Olive buff to brown
10.	Sabouraud's Agar	2.50-5.20 μ	Olive Buff	Septate	Dark brown	Terminal and intercalary	6.80-10.40 μ	22.70-62.30 \times 4.50-6.30 μ	Mid olive to brown

TABLE - XV (c)

Comparative study of cultural characters (size, colour, number of transverse and longitudinal septa of conidia and size, shape, colour and number of transverse septa of beak of conidia) of pathogen, *Alternaria alternata* (Fries.), Keissler, causing leaf spot disease of Dolichos bean (*Dolichos lablab*, L.) on different types of media.

S. No.	Media	Conidia					Beak			
		Size	Colour	Number of conidia	Number of transverse septa	Number of longitudinal septa	Size	Shape	Colour	Transverse septa
1.	Asthana and Hawker's Agar	7.0-23.0 × 3.60-7.40μ	Dark olive brown	2-5	1-6	0-3	1.0-4.80 × 1.60-4.80μ	Conical	Olive green	0-1
2.	Brown's Agar	7.0-24.60 × 3.40-7.40μ	Olive buff to brown	2-5	1-6	0-3	1.0-6.40 × 1.8-4.30μ	Conical	Olive buff to brown	0-2
3.	Coon's Agar	7.0-20.80 × 3.0-6.20μ	Light brown	2-5	2-7	0-3	1.0-3.0 × 1.30-3.40μ	Conical	Light brown	0-2
4.	Corn Meal Agar	7.0-23.40 × 3.50-6.40μ	Dark olive grey	2-6	1-4	0-3	1.80-73.60 × 1.40-3.60μ	Cylindrical	Olive grey	0-2
5.	Czapek's Agar	7.50-32.60 × 3.90-10.20μ	Olivaceous to dark brown	2-6	2-7	0-4	2.50-18.80 × 2.0-5.70μ	Cylindrical	Dark brown	0-1
6.	Leaf Decoction Agar	7.80-23.60 × 3.90-7.40μ	Deep olive brown	2-6	2-7	0-4	1.50-73.50 × 1.50-3.0μ	Cylindrical	Dark olive brown	0-1

7.	Malt Salt Agar	7.40-28.60 × 3.90-8.50μ	Olivaceous to dark brown	2-6	2-6	0-4	2.80-78.40 × 1.60-4.50μ	Conical	Olive to dark brown	0-1
8.	Potato Dextrose Agar	7.50-35.0 × 4.0-13.80μ	Olivaceous to brown	3-7	2-8	0-6	3.0-22.50 × 2.60-7.40μ	Conical	Light olive buff	0-2
9.	Richard's Agar	7.40-33.80 × 3.90-12.20μ	Olivaceous to dark brown	2-7	2-8	0-5	5.50-24.20 × 2.80-6.20μ	Cylindrical	Light brown	0-2
10.	Sabouraud's Agar	7.20-32.40 × 3.80-10.40μ	Olivaceous to dark brown	2-5	2-6	0-4	2.50-19.50 × 2.0-5.10μ	Cylindrical	Dark brown	0-1

Septation in conidia was also observed variable on different types of media. Transverse septa varied from 1-6 to 2-8 viz., 1-4 on Corn meal agar media; 1-6 on Asthana and Hawker's agar and Brown's agar media; 2-7 on Coon's agar; Czapek's agar; Leaf decoction agar; Malt salt agar and Sabouraud's agar media and 2-8 on Potato dextrose agar and Richard's agar media, while longitudinal septa varied from 0-3 to 0-6 viz., 0-3 on Asthana and Hawker's agar; Brown's agar; Coon's agar and Corn meal agar media; 0-4 on Czapek's agar, Leaf decoction agar; Malt salt agar and Sabouraud's agar media; 0-5 on Richard's agar medium and 0-6 on Potato dextrose agar medium. Conidia were also found varied in size from $7.0-35.0 \times 3.0-13.80 \mu$ on different types of media.

The beaks were found septate and varied in size from $1.0-78.40 \times 1.30-7.40 \mu$ on different types of media. The transverse septa varied from 0-2 viz., 0-1 on Asthana and Hawker's agar; Czapek's agar; Leaf decoction agar; Malt salt agar and Sabouraud's agar media and 0-2 on Brown's agar; Coon's agar; Corn Meal agar; Potato dextrose agar and Richard's agar media. The beaks were also observed varying in colour viz., olive green on Asthana and Hawker's agar medium; olive buff to brown on Brown's agar medium; light brown on Coon's agar and Richard's agar media; olive grey on Corn Meal agar medium; dark brown on Czapek's agar and Sabouraud's agar media; dark olive brown on leaf decoction agar medium; olive to dark brown on Malt salt agar medium and light olive buff on Potato dextrose agar medium. The beaks were found varying in shape viz., cylindrical on Corn Meal agar; Czapek's agar; Leaf decoction agar; Richard's agar and Sabouraud's agar media and conical on Asthana and Hawker's agar, Brown's agar, Coon's agar, Malt salt agar and Potato dextrose agar media.

The chlamydospores, were found terminal as well as intercalary varying in size from $4.70 - 22.80 \mu$ in diameter on all the different types of media under study but varying in colour viz., dark brown on Czapek's agar and Sabouraud's agar media; dark olive green on Asthana and Hawker's agar medium; dark olive brown on leaf decoction agar medium; olivaceous to dark brown on Malt salt

agar medium; olive buff on Potato dextrose agar medium; light brown on Coon's agar and Richard's agar media and olive grey on Corn Meal agar medium.

GROWTH OF THE PATHOGEN ON LIQUID MEDIA :

For the study of growth, the pathogen was grown on non-synthetic, semi-synthetic and synthetic media and observations were recorded on average fungal dry weight; sporulation and other characters of the pathogen after 10 days of incubation at $25\pm 1^{\circ}\text{C}$ according to the method described in "Material and Method" as summarised in Table XVI and Figures 15 and 16.

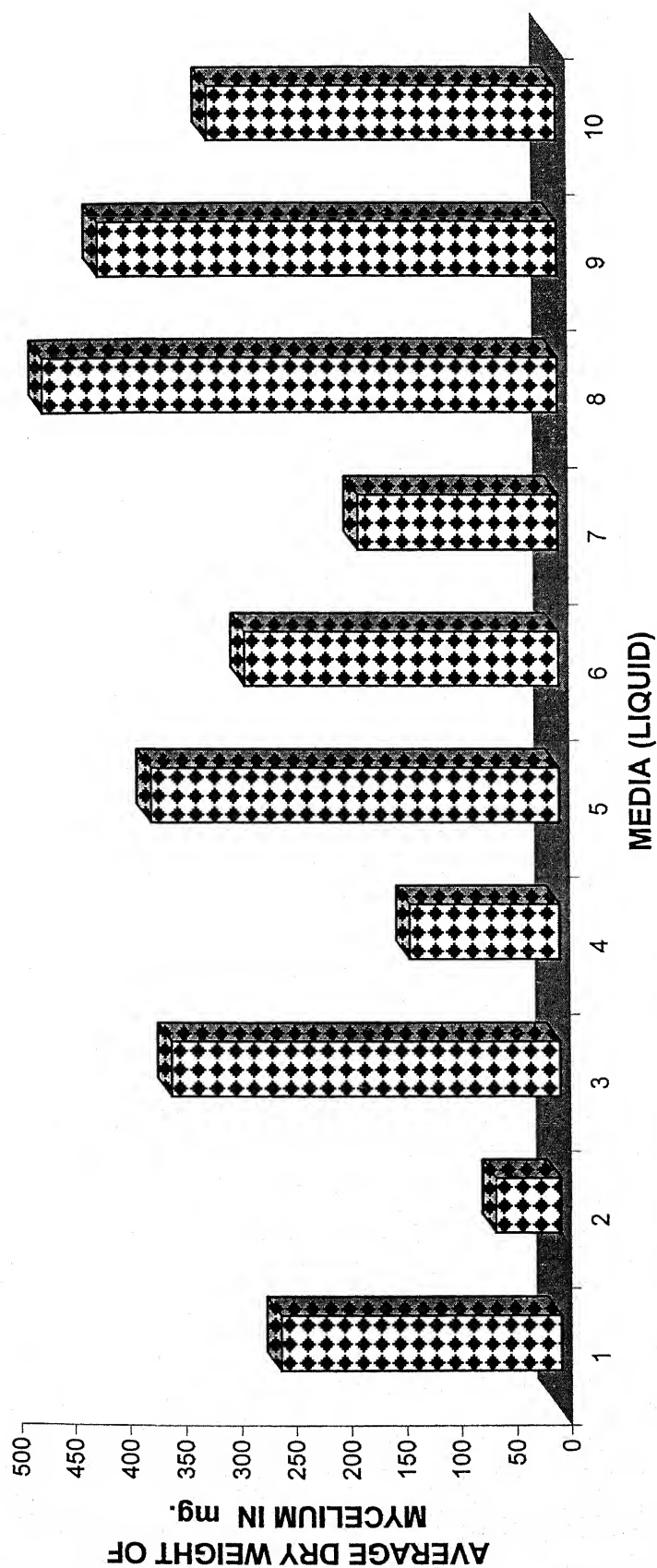
TABLE - XVI

Average fungal dry weight and sporulation of *Alternaria alternata* (Fries.), Keissler, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.) on different liquid media at $25\pm 1^{\circ}\text{C}$ after 10 days of incubation.

S.No.	Media (Liquid)	Average dry weight of Fungus (mg.)	Sporulation
1.	Asthana and Hawker's medium	254.60	+
2.	Brown's medium	58.70	+
3.	Coon's medium	352.50	+++
4.	Corn Meal medium	136.00	+
5.	Czapek's medium	370.20	++++
6.	Leaf Decoction medium	285.30	+
7.	Malt Salt medium	182.00	+
8.	Potato dextrose medium	467.40	++++
9.	Richard's medium	416.50	++++
10.	Sabouraud's medium	317.00	+++

- (+) = Denotes Poor sporulation.
 (++) = Denotes Fair sporulation.
 (+++) = Denotes Good sporulation.
 (++++) = Denotes Excellent sporulation.

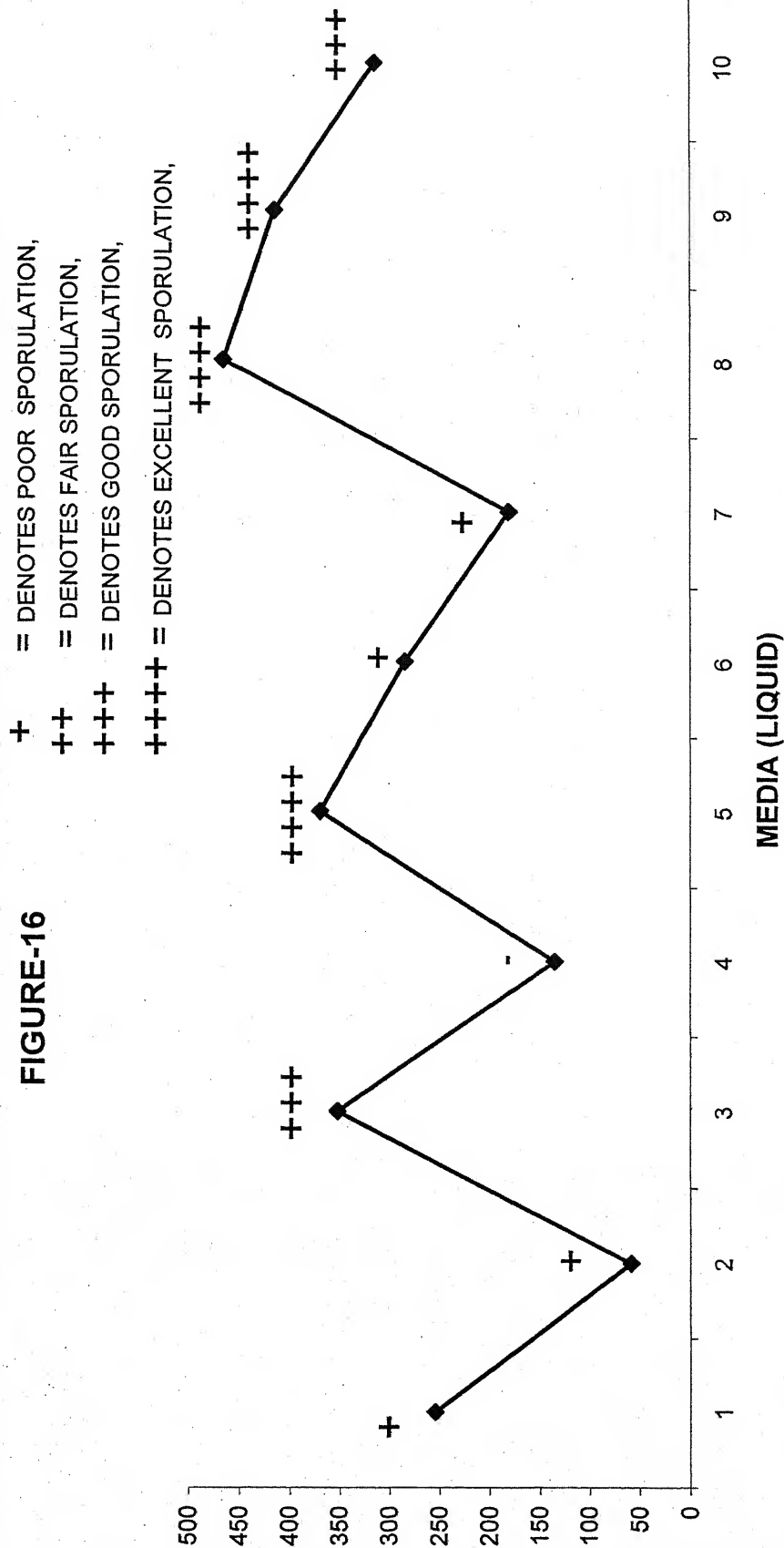
FIGURE-15



EFFECT OF DIFFERENT LIQUID MEDIA ON GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler.

FIGURE-16

AVERAGE DRY WEIGHT OF MYCELIIUM IN mg.



EFFECT OF DIFFERENT LIQUID MEDIA ON GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler.

Data presented in Table XVI and corresponding Figures 15 and 16 revealed that all the media tested differed significantly. The maximum growth of pathogen, weighing 467.40 mg. was obtained on Potato dextrose medium followed by Richard's medium weighing 416.50 mg. A moderately good growth was supported on Czapek's; Coon's and Sabouraud's media, while Asthana and Hawker's; Leaf decoction and Malt salt media exhibited fair growth. Brown's and Corn Meal media showed poor growth. The minimum growth and minimum dry weight of fungal mycelium weighing 58.70 mg. was observed on Brown's medium.

Czapek's, Potato dextrose and Richard's media proved to be excellent in sporulation, while Coon's and Sabouraud's media supported good sporulation. Fair sporulation, was exhibited on Asthana and Hawker's; leaf decoction and Malt salt media, while poor on Brown's and Corn Meal media.

SELECTION OF BASAL MEDIUM :

It is obvious from the findings recorded that Potato dextrose agar medium is superior to all other non-synthetic, semi-synthetic and synthetic media. The Potato dextrose agar medium supported best mycelial growth and excellent sporulation of the pathogen and contains almost all the elements required for the best growth of pathogen, *Alternaria alternata* (Fries.), Keissler. Thus Potato dextrose agar medium, was selected as a basal medium for further studies.

PHYSIOLOGICAL STUDIES :

(a) EFFECT OF TEMPERATURE OF GROWTH AND SPORULATION OF THE PATHOGEN :

To the study the effect of temperature on growth and sporulation the pathogen was grown in flasks on Potato dextrose agar liquid medium and incubated for 10 days at different temperatures, viz., 5°C (T₁), 10°C (T₂), 15°C (T₃), 20°C (T₄), 25°C (T₅), 30°C (T₆), 35°C (T₇), 40°C (T₈), 45°C (T₉), and 50°C

(T₁₀), according to the method described under, "Material and Method". The observations regarding the average fungal dry weight and extent of sporulation of pathogen were recorded as expressed in Table XVII and Figures 17 and 18.

TABLE - XVII

Average Fungal dry weight and sporulation of *Alternaria alternata* (Fries.), Keissler, causing leaf Spot of Dolichos bean (*Dolichos lablab*, L.) on Potato Dextrose liquid media at different temperatures after 10 days of incubation.

S. No.	Temperature (°C)	Average Fungal Dry weight (mg.)	Sporulation
1.	5 (T ₁)	87.60	—
2.	10 (T ₂)	143.80	+
3.	15 (T ₃)	216.40	+
4.	20 (T ₄)	312.00	+++
5.	25 (T ₅)	381.70	++++
6.	30 (T ₆)	461.20	++++
7.	35 (T ₇)	435.00	++++
8.	40 (T ₈)	347.40	+++
9.	45 (T ₉)	175.00	+
10.	50 (T ₁₀)	95.20	—

(-) = Denotes absence of sporulation.

(+) = Denotes poor sporulation.

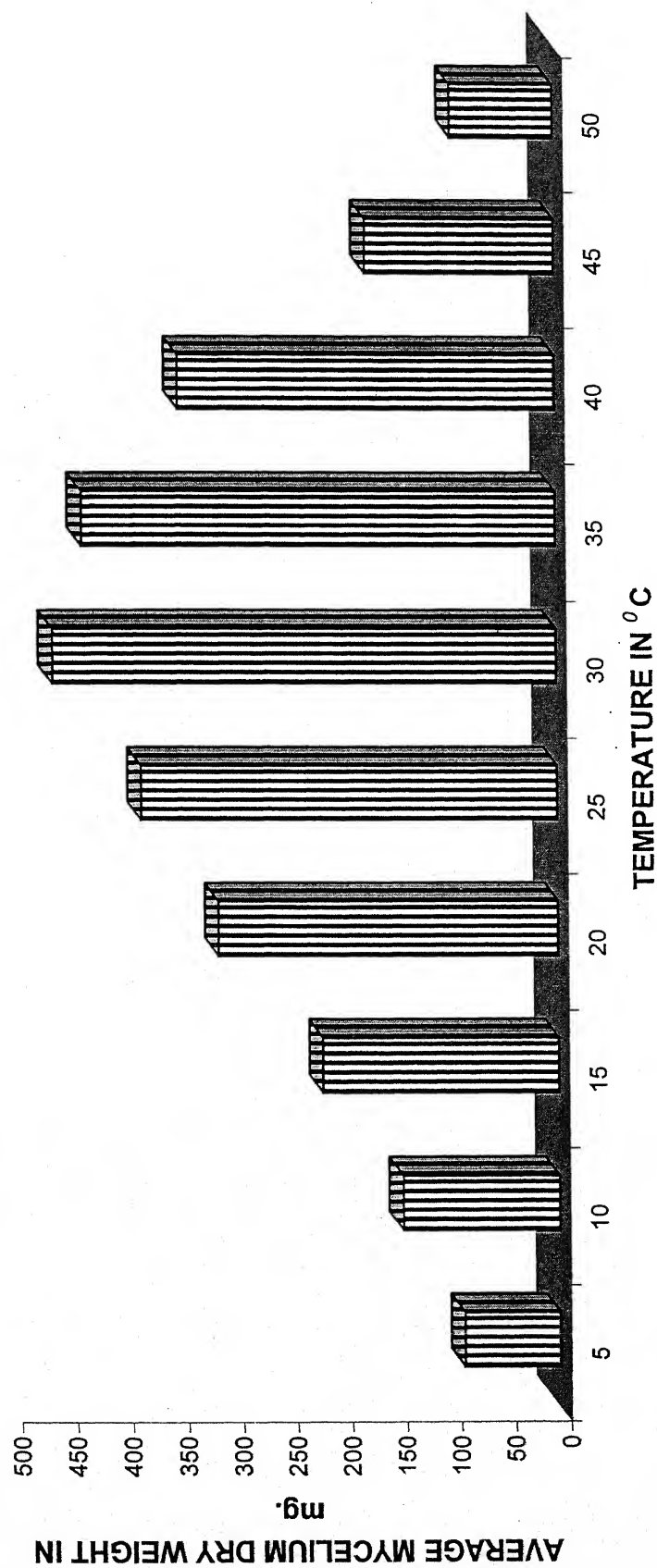
(++) = Denotes fair sporulation.

(+++)= Denotes good sporulation.

(++++)= Denotes excellent sporulation.

Data presented in Table XVII and corresponding Figures 17 and 18, exhibited that the pathogen grew in culture at wide range of temperatures viz.,

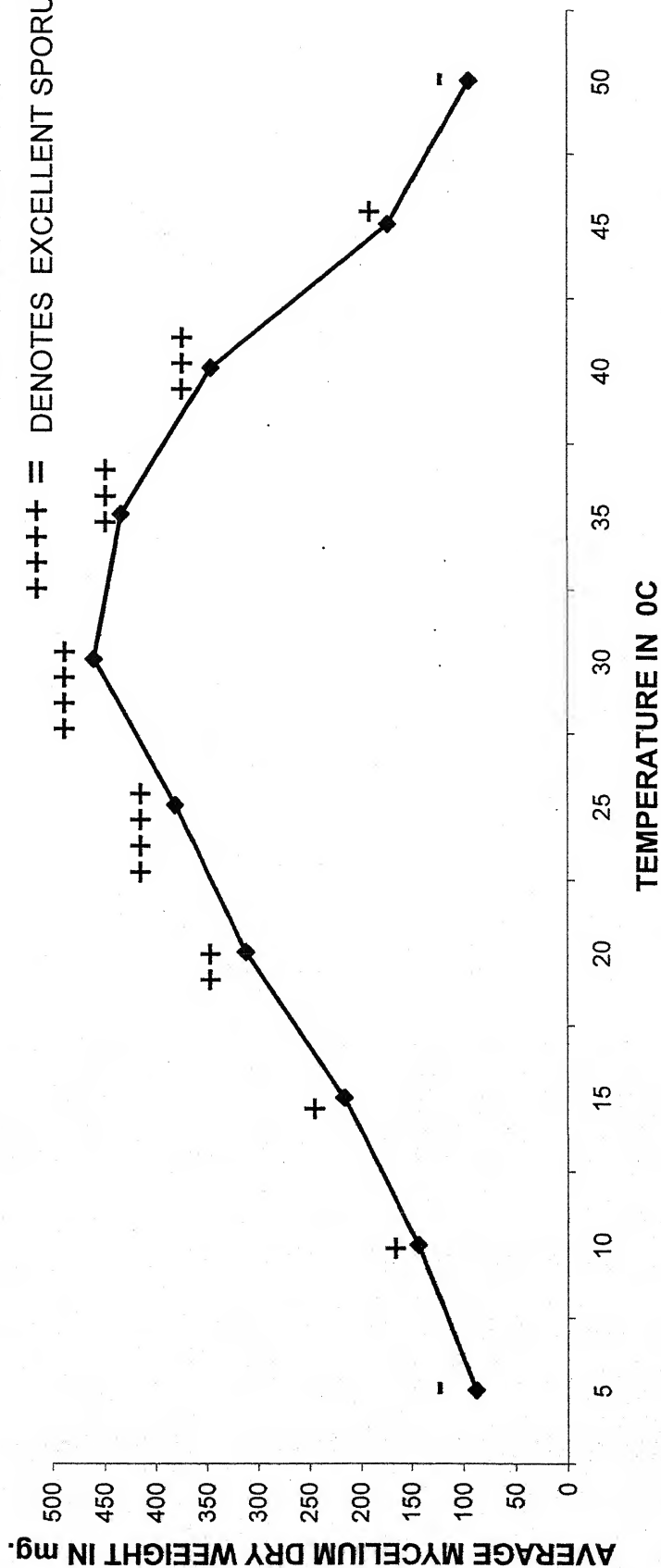
FIGURE-17



EFFECT OF DIFFERENT TEMPERATURES ON GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler.

FIGURE-18

- = DENOTES ABSENCE OF SPORULATION.
 + = DENOTES POOR SPORULATION.
 ++ = DENOTES FAIR SPORULATION.
 +++ = DENOTES GOOD SPORULATION.
 ++++ = DENOTES EXCELLENT SPORULATION.



EFFECT OF DIFFERENT TEMPERATURES ON GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler.

5°C (T₁), 10°C (T₂), 15°C (T₃), 20°C (T₄), 25°C (T₅), 30°C (T₆), 35°C (T₇), 40°C (T₈), 45°C (T₉), and 50°C (T₁₀), expressed significant differences in fungal dry weight. The maximum growth of the pathogen, was obtained at 30°C (T₆), having a dry weight of 461.20 mg., followed by 35°C having a dry weight of 435.0 mg. Other higher temperatures viz., 40°C (T₈), 45°C (T₉) and 50°C (T₁₀), reduced the growth to a great extent in comparison to lower temperatures viz., 5°C (T₁), 10°C (T₂), 15°C (T₃), 20°C (T₄) and 25°C (T₅), where as the minimum fungal growth was observed having a dry weight of 87.60 mg. at 5°C (T₁). Thus it is evident that pathogen grew well at 25°C (T₅), 30°C (T₆) and 35°C (T₇) but the optimum temperatures range was 30°C (T₆) and 35°C (T₇).

The optimum temperature for sporulation proved closely related to the growth of pathogen. The best sporulation, was recorded at 30°C (T₆); excellent sporulation at 25°C (T₅) and 35°C (T₇); good at 20°C (T₄) and 40°C (T₈); while fair at 15°C (T₃) and 45°C (T₉) and poor at 10°C (T₂). The pathogen established as failed to sporulate at minimum and maximum temperature respectively 5°C (T₁) and 50°C (T₁₀), the minimum and maximum temperatures under investigation.

EFFECT OF HYDROGEN-ION CONCENTRATION (pH) ON THE GROWTH AND SPORULATION OF PATHOGEN :-

The hydrogen-ion (pH) concentration of the medium causes significant and pronounced effect on the growth and sporulation of fungus grown in 20 different pH values Viz., P1 (pH 2.50), P2 (pH 3.0), P3 (pH 3.50), P4 (pH 4.0), P5 (pH 4.50), P6 (pH 5.0), P7 (pH 5.50), P8 (pH 6.0), P9 (pH 6.50), P10 (pH 7.0), P11 (pH 7.50), P12 (pH 8.0), P13 (pH 8.50), P14 (pH 9.0), P15 (pH 9.50), P16 (pH 10.0), P17 (pH 10.50), P18 (pH 11.0), P19 (pH 11.50) and P20 (pH 12.0), after 10 days of incubation at 25±1°C according to the method described under "Material and Method". The observations regarding the average fungal dry weight and extent

of sporulation of pathogen, were noted as detailed in Table XVIII and Figures 19 and 20.

TABLE - XVIII

Average Fungal growth and sporulation of pathogen *Alternaria alternata* (Fries.), Keissler, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.) in Potato dextrose liquid medium adjusted at pH values after 10 days of incubation.

S.No.	Initial pH (mg.)	Fungal Dry weight (mg.)	pH after the growth of fungus	Sporulation
1.	2.50 (P 1)	71.70	6.30	—
2.	3.0 (P 2)	115.40	6.50	—
3.	3.50 (P 3)	168.20	7.60	+
4.	4.0 (P 4)	272.50	7.60	+
5.	4.50 (P 5)	303.60	7.10	+
6.	5.0 (P 6)	342.20	7.40	+
7.	5.50 (P 7)	385.00	7.40	+
8.	6.0 (P 8)	433.10	—	+
9.	6.50 (P 9)	495.00	7.50	++++
10.	7.0 (P 10)	460.70	7.50	++++
11.	7.50 (P 11)	402.50	—	+++
12.	8.0 (P 12)	325.30	8.20	+++
13.	8.50 (P 13)	287.50	8.50	+
14.	9.0 (P 14)	225.30	8.50	+
15.	9.50 (P 15)	102.40	8.90	+

16.	10.0 (P 16)	67.50	8.70	+
17.	10.50 (P 17)	58.00	8.30	+
18.	11.0 (P 18)	55.00	8.10	-
19.	11.50 (P 19)	57.10	7.50	-
20.	12.0 (P 20)	53.00	7.20	-

(-) = Denotes Absence of sporulation.

(+) = Denotes Poor sporulation.

(++) = Denotes Fair sporulation.

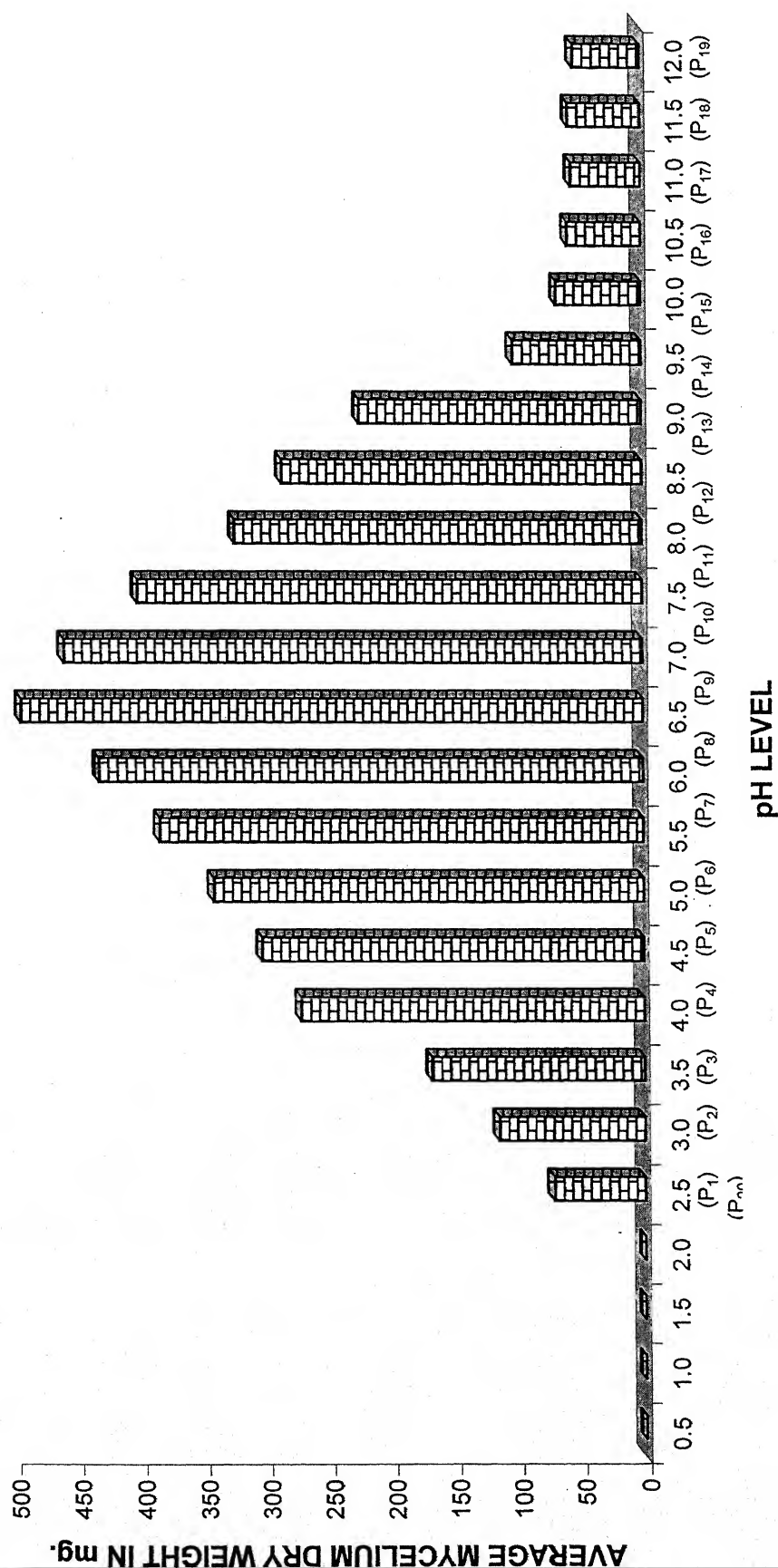
(+++)= Denotes Good sporulation.

(++++)= Denotes Excellent sporulation.

It is evident from the table XVIII and corresponding Figures 19 and 20 that the pathogen, *Alternaria alternata* (Fries.). Keissler, grew exhibiting significant differences in fungal growth at different pH values varying from 2.50-12.0. The maximum growth weighing 495.0 mg. was obtained at P9 (pH 6.50), followed by P10 (7.0) and P8 (6.0), weighing 460.70 mg. and 433.10 mg. respectively. At high pH values viz., P11 (pH 7.50), P12 (pH 8.0), P13 (pH 8.50), P14 (pH 9.0), P15 (pH 9.50), P16 (pH 10.0), P17 (pH 10.50), P18 (pH 11.0), P19 (pH 11.50) and P20 (pH 12.0), the growth was reduced gradually from 402.50 mg to 53.00 mg. The least growth weighing 71.70 mg. and 53.00 mg. were recorded at P1 (pH 2.50) and P20 (pH 12.0), which suggest that high alkaline and acidic pH did not favoured the growth of pathogen.

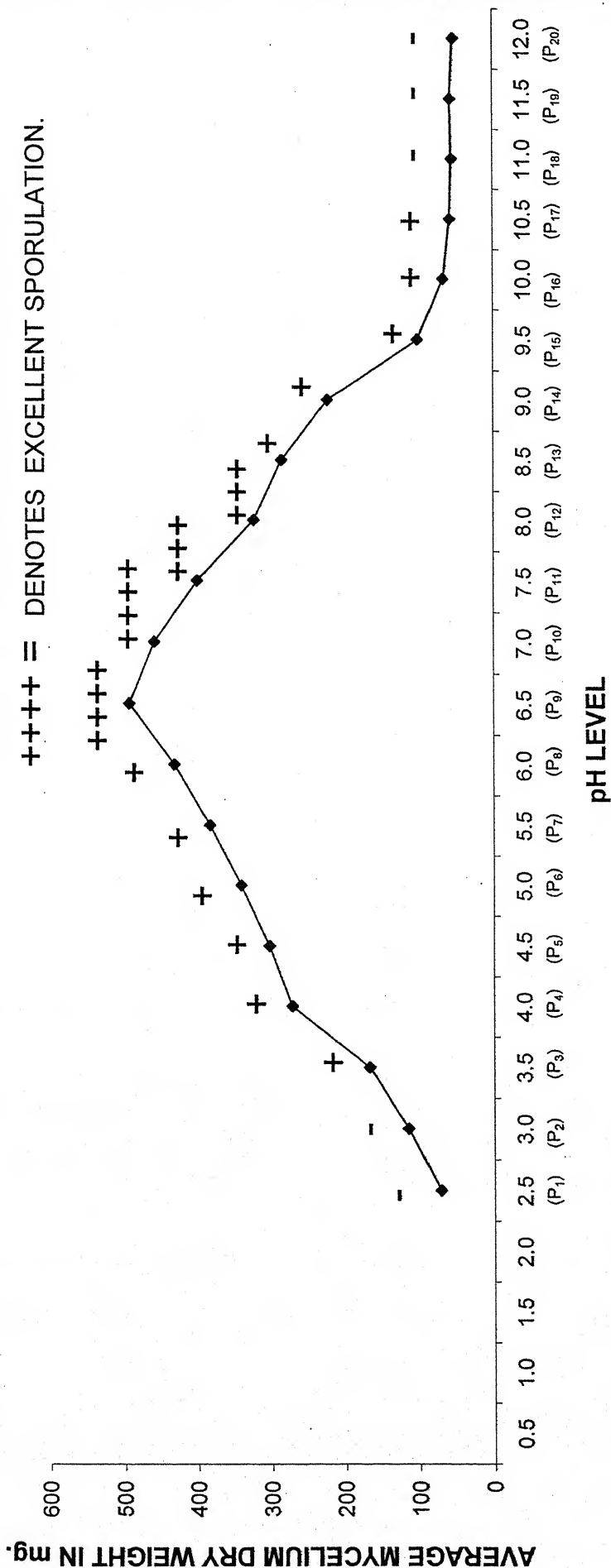
Excellent sporulation exhibited by the pathogen, was obtained at P9 (pH 6.50), which correspond highest fungal growth. Maximum sporulation recorded at P9 (pH 6.50), was significant being superior to all other pH values followed by P10 (pH 7.0). The other pH values viz., P11 (pH 7.50) and P12 (pH 8.0) also favoured good sporulation. Sporulation was recorded fair at P5 (pH 4.50), P6 (pH 5.0), P7 (pH 5.50), P8 (pH 6.0), P13 (pH 8.50), P14 (9.0) and P15 (9.50),

FIGURE-19



EFFECT OF DIFFERENT HYDROGEN-ION CONCENTRATION ON GROWTH AND SPORULATION OF
Alternaria alternata (Fries.), Keissler.

FIGURE-20



EFFECT OF DIFFERENT HYDROGEN-ION CONCENTRATION ON GROWTH AND SPORULATION OF
Alternaria alternata (Fries.), Keissler.

whereas P3 (pH 3.50), P4 (pH 4.0), P16 (pH 10.0) and P17 (pH 10.50) showed poor sporulation. However the pathogen failed to sporulate on P1 (pH 2.50), P2 (pH 3.0), P18 (pH 11.0), P19 (pH 11.50) and P20 (pH 12.0).

After culturing the pathogen, it was also observed that reaction of medium tended towards alkaline in cases, where the initial pH was on acidic side and vice versa in the cases, where the media was adjusted at P14 (pH 9.0) to P16 (pH 10.0) initially.

PRODUCTION OF CELLULOLYTIC AND PECTINOLYTIC ENZYMES BY THE PATHOGEN *Alternaria alternata* (Fries.). Keissler, In Vitro.

In general degradation of cell wall components is essential for the successful invasion of the host tissues by the phytopathogenic micro-organisms. The pathogenic fungi produce cell wall degrading enzymes in *vivo* and *vitro* during growth upon which infection depends. The structural framework of plant cell is largely composed of cellulosic and pectic substances. Taking into consideration the following experiments were conducted to detect the production of Cellulolytic and Pectinolytic enzymes by the pathogen in *vivo* and *vitro*.

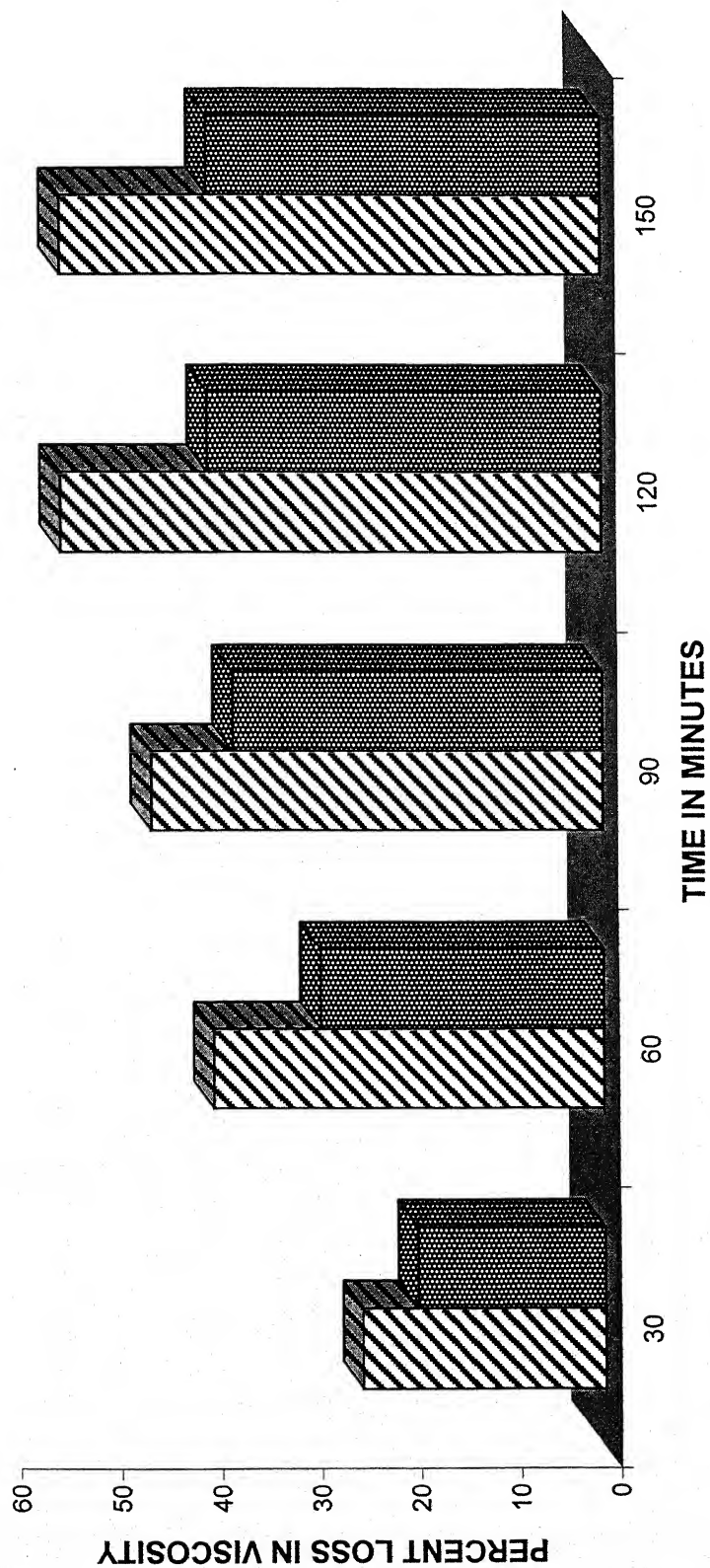
PRODUCTION OF ENZYMES IN VITRO :

The pathogen, was grown on Richard's liquid medium and the activity of Cellulase (CX), Polymethylgalacturonase (PMG) and Polygalacturonase (PG) enzymes produced by the pathogen, *Alternaria alternata* (Fries.), Keissler, was determined as described under, "Material and Method". The enzymatic activity was expressed in terms of per cent loss of viscosity of the substrates over control after giving a reaction time as presented in Tables XIX, XX and XXI and Figures 21, 22, 23, 24, 25 and 26.

1. CELLULASE (CX) ASSAY :

The production and activity of Cellulase (Cx) produced by the culture was determined by Viscometric method . The result of production and activity of Cellulase (Cx) enzymes secreted by the pathogen, *Alternaria alternata* (Fries.) Keissler, are given in table XIX and Figures 21 and 22.

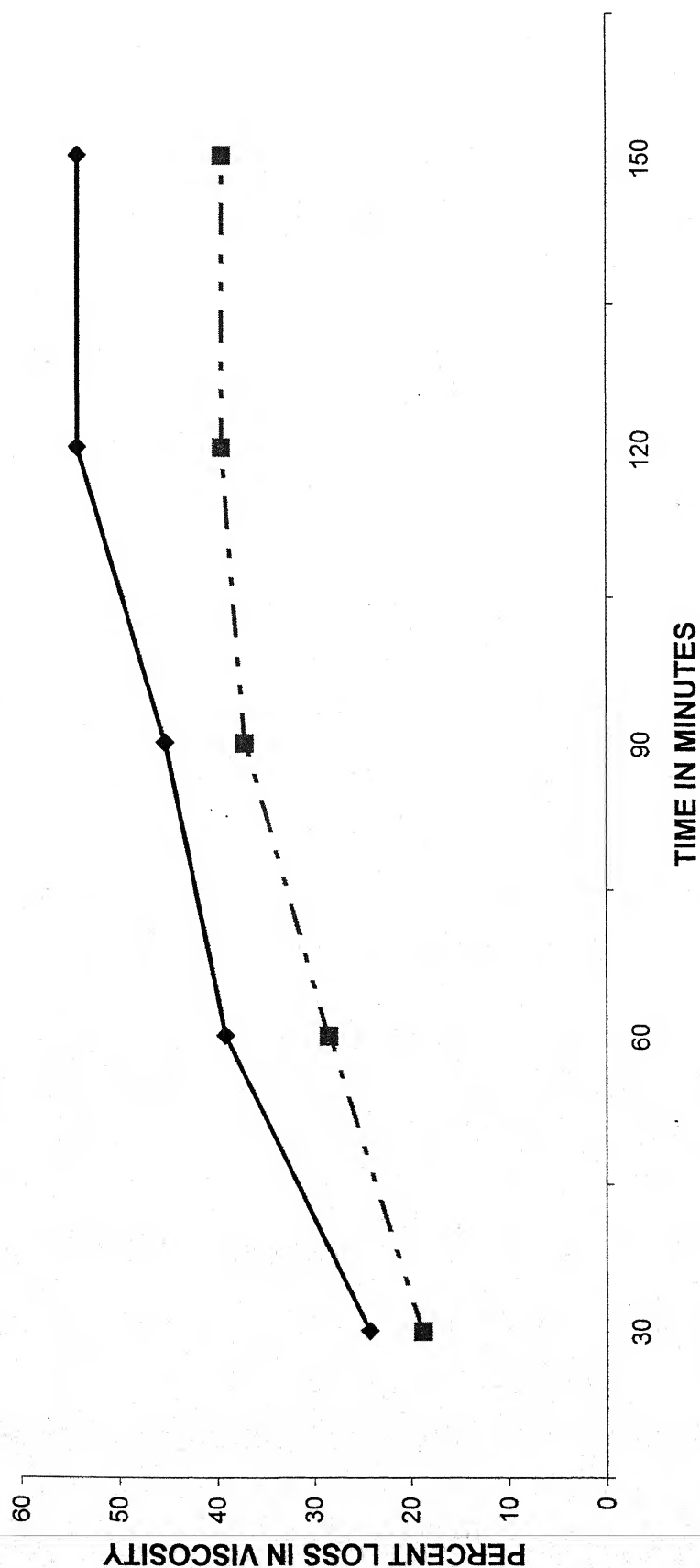
FIGURE-21



PRODUCTION AND ACTIVITY OF CELLULASE (CX) EZYME BY *Alternaria alternata* (Fries.), Keissler. IN CULTURE MEDIUM IN VITRO.

FIGURE-22

—◆— WITH CARBOXYMETHYL CELLULOSE
—■— WITHOUT CARBOXYMETHYL CELLULOSE



PRODUCTION AND ACTIVITY OF CELLULASE (CX) ENZYME BY *Alternaria alternata* (Fries.), Keissler. IN CULTURE MEDIUM IN VITRO.

TABLE - XIX

Production and Activity of Cellulase (CX) enzyme by *Alternaria alternata* (Fries.), Keissler, in culture medium in *Vitro.*, Causing leaf spot of Dolichos bean (*Dolichos lablab*, L.)

Treatments	Percent Loss of Viscosity at time intervals (minutes)				
	30	60	90	120	150
With carboxymethyl cellulose (CMC)	24.30	39.20	45.37	54.28	54.28
Without Carboxymethyl cellulose (CMC)	18.75	28.50	37.20	39.64	39.64

From the data given in Table XIX and corresponding Figures 21 and 22, exhibited that production and activity of Cellulase (Cx) enzyme, was greatly influenced by the medium used. The pathogen produced Cellulase (Cx) enzyme in culture filterates of medium with and without Carboxymethyl cellulose (CMC), but its production and activity was comparatively much more in the medium supplemented with Carboxymethyl cellulase (CMC) as compared to that devoid of it. The per cent loss in viscosity increased with time and became constant after 120 minutes in the medium with and without Carboxymethyl cellulose (CMC). Highest activity of Cellulase (Cx), was observed after 120 minutes of incubation at 54.28 per cent loss of viscosity of enzymes sample with substrate. The medium, which was without substrate produced less amount of enzyme as 39.64 per cent loss of viscosity in the same period of incubation.

2. POLYGALACTURONASE (PG)

The production and activity of Polygalacturonase (PG) by the pathogen *Alternaria alternata*, was determined by Viscometric method and the results are given in Table XX and Figures 23 and 24.

TABLE - XX

Production and Activity of Polygalacturonase (PG) enzyme by *Alternaria alternata* (Fries.), Keissler, in culture medium in *Vitro.*, causing *Alternaria* leaf spot of *Dolichos* bean (*Dolichos lablab*, L.)

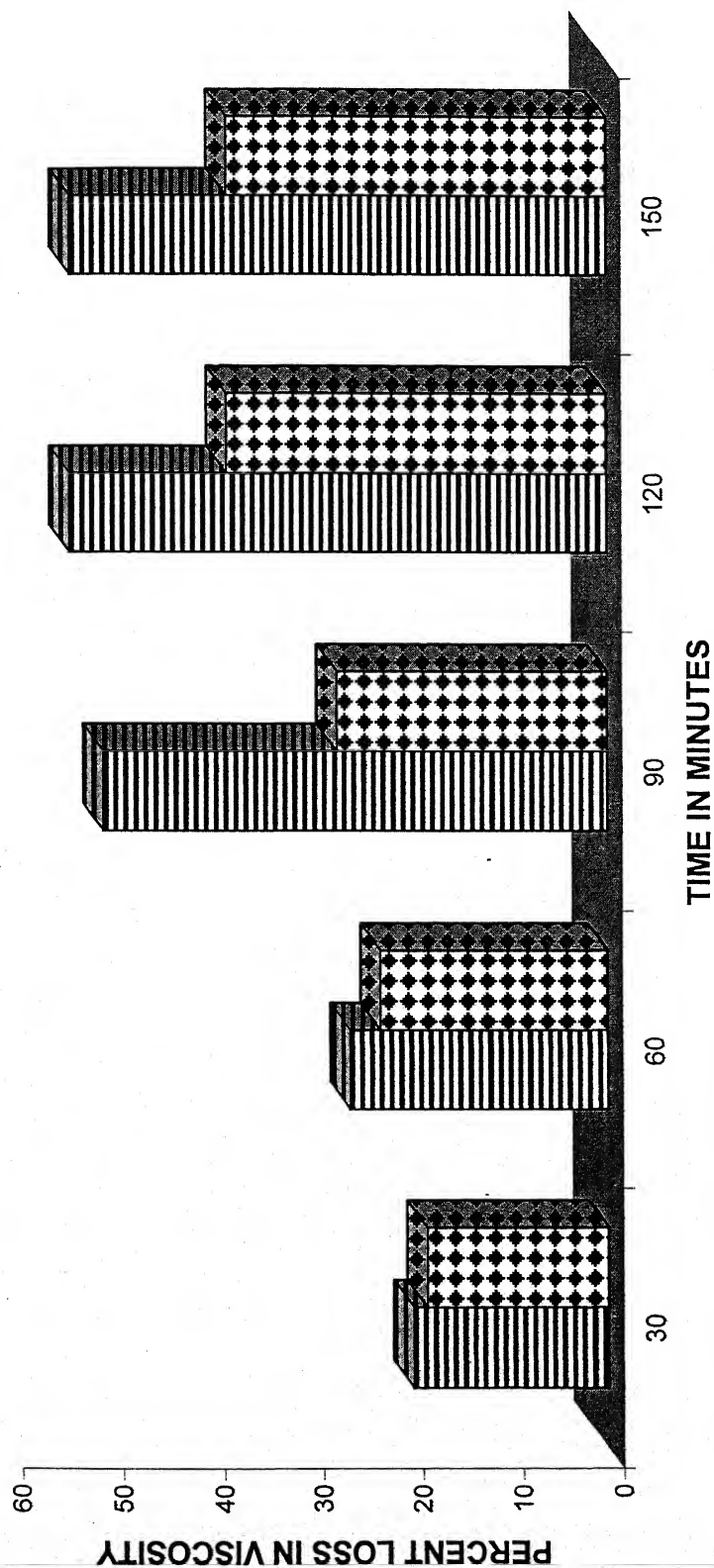
Treatments	Percent Loss of Viscosity at time intervals (minutes)				
	30	60	90	120	150
With Sodium Polypectate	19.45	25.80	50.40	53.68	53.68
Without Sodium Polypectate	18.10	22.73	27.15	38.20	38.20

The results obtained in Table XX and its corresponding Figures 23 and 24, revealed that the pathogen, *Alternaria alternata* (Fries.), Keissler, produced more amount of Polygalacturonase (PG) enzyme in the medium supplemented with sodium polypectate. In the medium with sodium polypectate the higher values of per cent loss in viscosity, were recorded in comparison to values recorded at the same time of interval in the medium without sodium polypectate. The per cent loss in viscosity increased gradually with the increase in time and become constant after 120 minutes of reaction in the medium with or without sodium polypectate. Highest activity of Polygalacturonase (PG) enzyme, was observed after 120 minutes of incubation as 53.68 per cent of loss viscosity of enzyme sample with substrate. The medium, which was without substrate produced lesser amount of enzyme as 38.20 per cent loss of viscosity of enzyme sample with substrate in the same period of incubation.

3. POLYMETHYLGALACTURONASE (PMG)

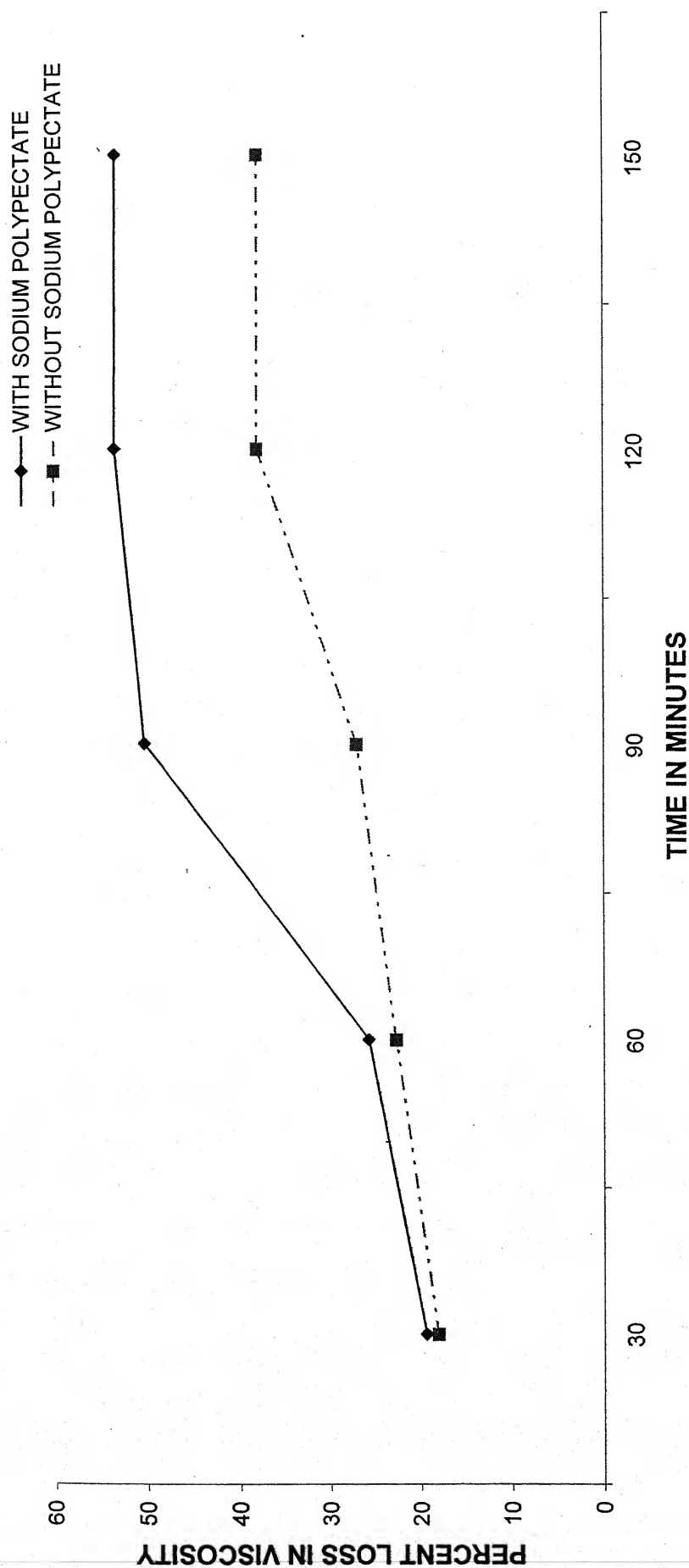
The activity and production of Polymethylgalacturonase (PMG) enzyme produced by the pathogen, *Alternaria alternata* (Fries.), Keissler, was determined by Viscometric method as discussed in "Material and Method". The technique adopted was the same as in case of Polygalacturonase (PG) assay except, Citrus pectin was taken in the place of sodium polypectate. The results in

FIGURE -23



PRODUCTION AND ACTIVITY OF POLYGALACTURONASE (PG) ENZYME BY *Alternaria alternata* (Fries.), Keissler. IN CULTURE MEDIUM IN VITRO.

FIGURE-24



PRODUCTION AND ACTIVITY OF POLYGALACTURONASE (PG) ENZYME BY *Alternaria alternata* (Fries.), Keissler, IN CULTURE MEDIUM IN VITRO.

loss of viscosity over control at different time periods recorded are given in the Table XXI and corresponding Figures 25 and 26.

TABLE - XXI

Production and Activity of Polymethylgalacturonase (PMG) enzyme by *Alternaria alternata* (Fries.), Keissler, in culture medium in *Vitro.*, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.)

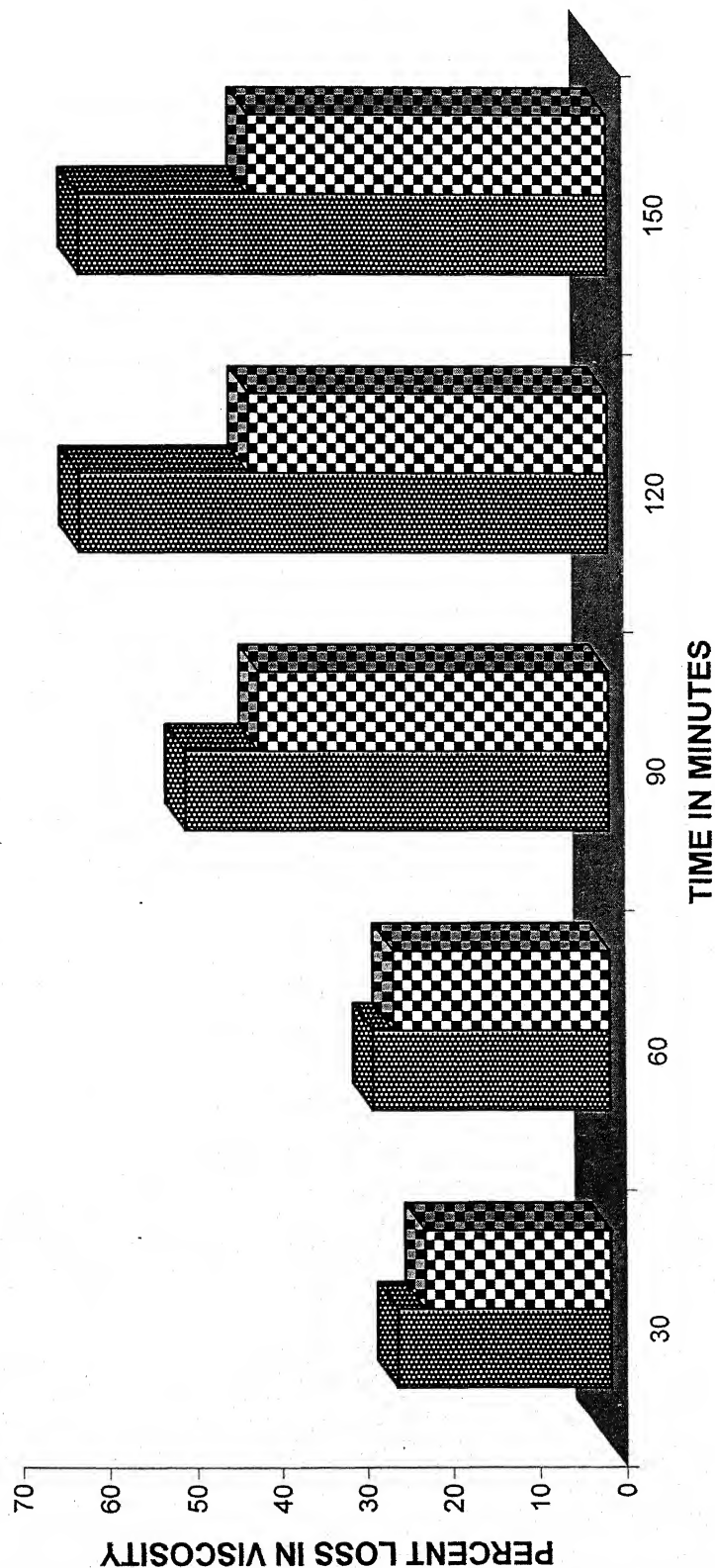
Treatments	Percent Loss of Viscosity at time intervals (minutes)				
	30	60	90	120	150
With Citrus pectin	24.78	27.55	49.37	61.38	61.38
Without Citrus pectin	21.54	25.20	40.85	41.90	41.90

From the data presented in Table XXI and its corresponding Figures 25 and 26, it is obvious that the pathogen, *Alternaria alternata* (Fries.), Keissler, produced polymethylgalacturonase (PMG) enzyme in the medium supplemented with and without pectin. The production of polymethylgalacturonase (PMG), was more in the medium supplemented with pectin as compared to the medium without pectin. The pathogen grown in the medium substituted with or without pectin increased with the increase in time intervals upto 120 minutes of standing and after that became constant. Further increase in the time did not increase the enzyme activity, which was constant after 120 minutes. Highest activity of Polymethylgalacturonase (PMG) enzyme, was observed after 120 minutes of incubation as 61.38 per cent loss of viscosity of enzyme sample with substrate. The medium, which was without substrate lesser amount of enzyme was observed as 41.90 per cent loss of viscosity of enzyme sample with substrate in the same period of incubation.

PRODUCTION OF ENZYMES IN VIVO :

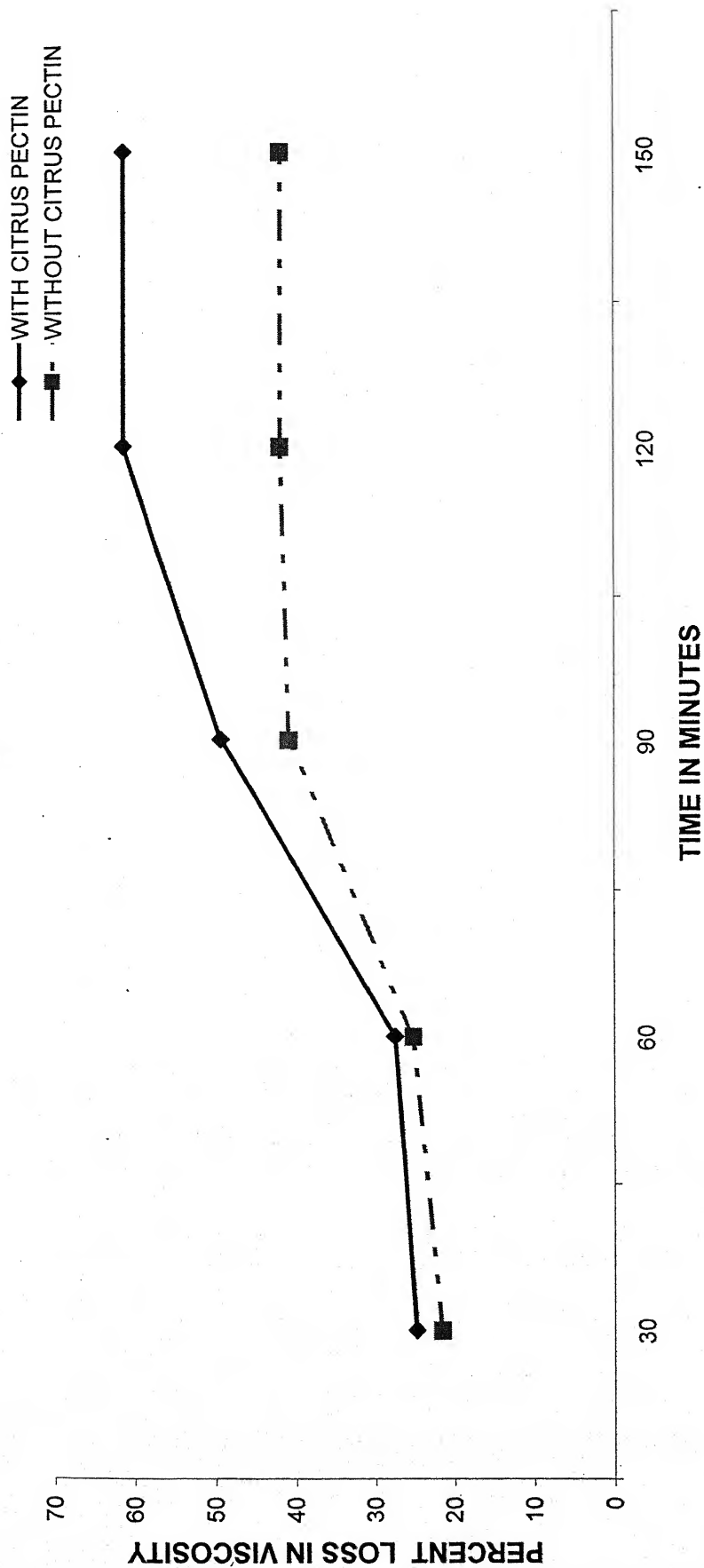
For the study of Cellulase (Cx), Polymethylgalacturonase (PMG) and Polygalacturonase enzyme (PG) in *vivo*, the extract of diseased leaves was used in place of culture filtrates as in *vitro*. The diseased leaves, were obtained by

FIGURE-25



PRODUCTION AND ACTIVITY OF POLYMETHYL GALACTURONASE (PMG) ENZYME BY *Alternaria alternata* (Fries.), Keissler. IN CULTURE MEDIUM IN VITRO.

FIGURE-26



PRODUCTION AND ACTIVITY OF POLYMETHYL GALACTURONASE (PMG) ENZYME BY *Alternaria alternata* (Fries.), Keissler. IN CULTURE MEDIUM IN VITRO.

inoculating the leaves of germplasm/culture "Kalyanpur Type-1" of Dolichos bean (*Dolichos lablab*, L.) by culture of *Alternaria alternata* (Fries.), Keissler, according to the technique described under "Material and Method". For comparison the extracts were also prepared from the healthy leaves of test germplasm/culture. The technique adopted was the same as in *vitro* studies. The results in terms of per cent loss of viscosity of the substrate are summarised in Table XXII, XXIII and XXIV and corresponding Figures 27, 28, 29, 30, 31 and 32 as in *vitro* studies. The maximum loss of viscosity, was observed after 120 minutes in Polymethylgalacturonase (PMG) and Polygalacturonase (PG) except in Cellulase (Cx) enzyme, where it was observed after 150 minutes.

1. CELLULASE (CX) ASSAY

The production and activity of Cellulase (Cx) enzyme produced by the pathogen *Alternaria alternata* (Fries.), Keissler, was determined by Viscometric method. The results of the production and activity of cellulase (Cx) enzymes secreted by pathogen are given in table XXII and corresponding Figures 27 and 28.

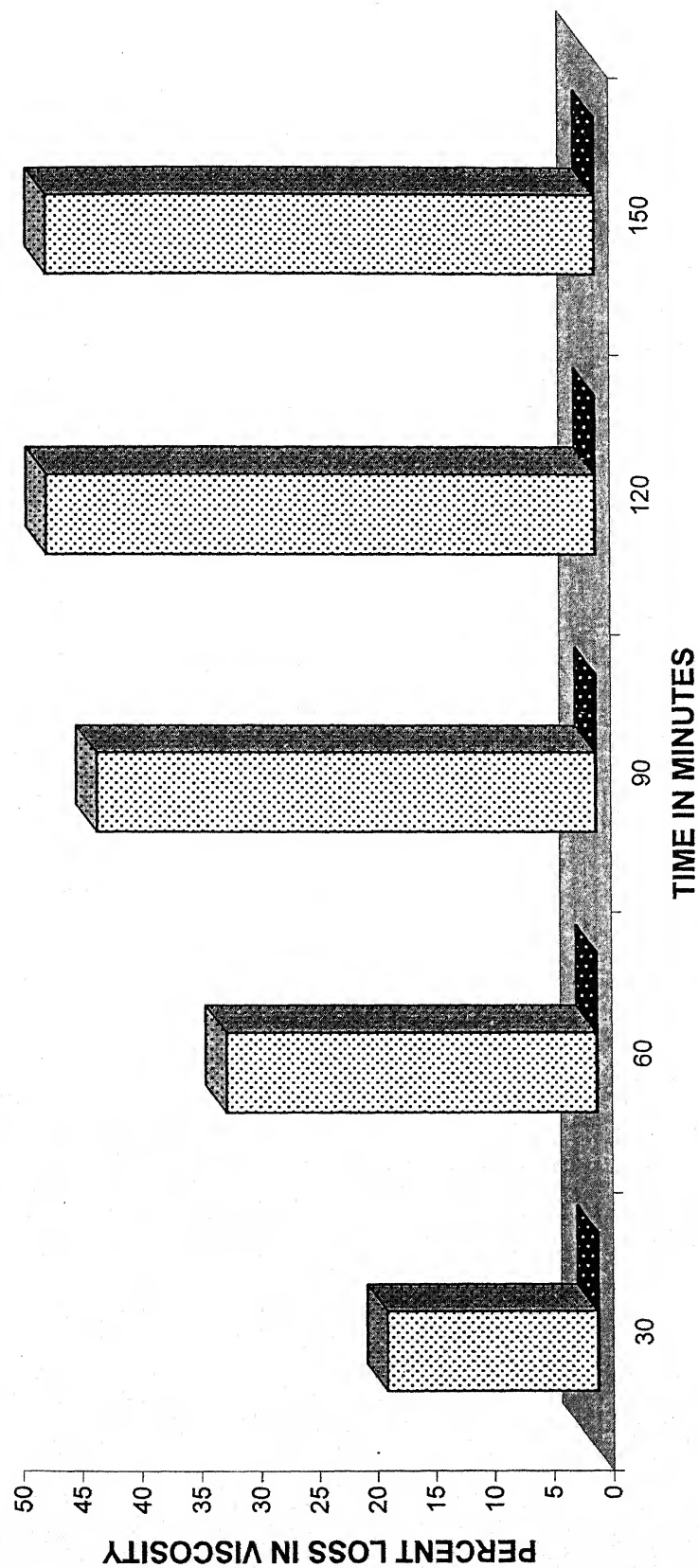
TABLE - XXII

Production and Activity of Cellulase (CX) enzyme by *Alternaria alternata* (Fries.), Keissler, in *Vivo.*, causing *Alternaria* leaf spot of Dolichos bean (*Dolichos lablab*, L.)

Treatments	Percent Loss of Viscosity at time intervals (minutes)				
	30	60	90	120	150
Diseased	17.85	31.57	42.35	46.50	46.50
Healthy	0.0	0.0	0.0	0.0	0.0

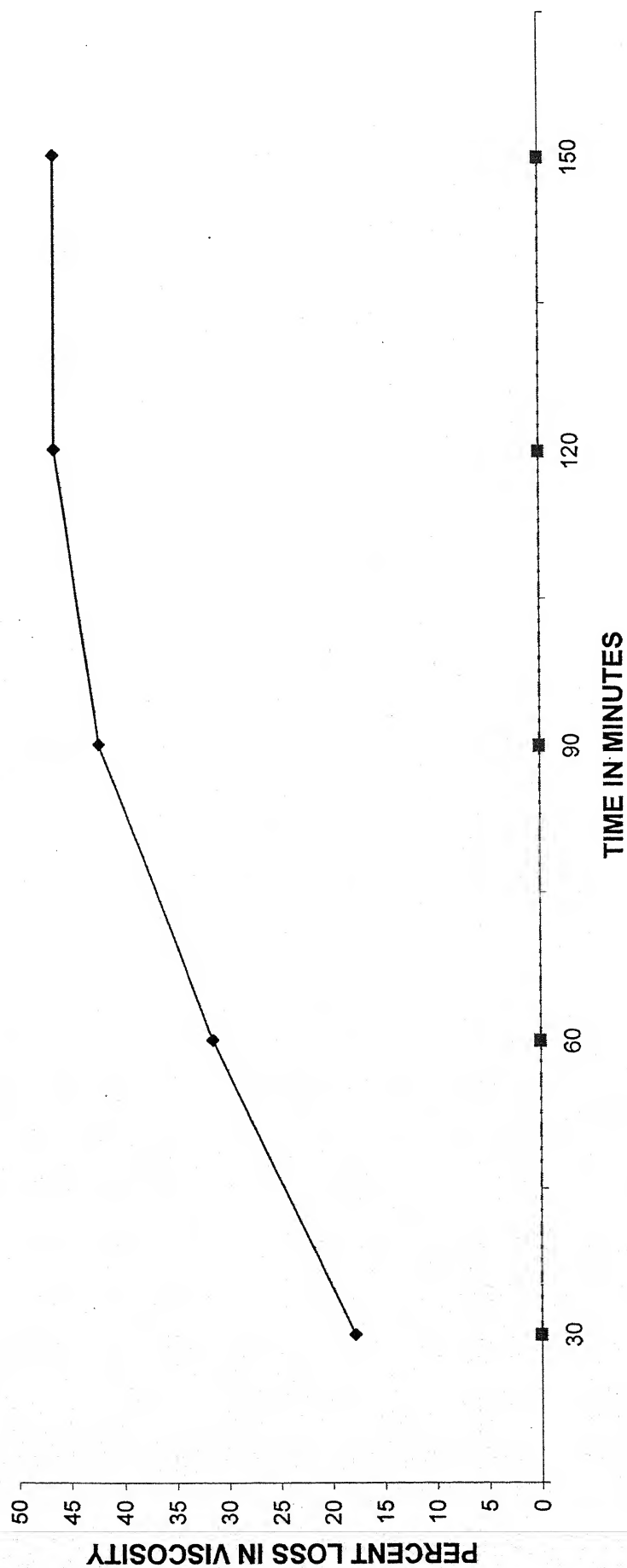
It is obvious from the data presented in Table XXII and corresponding Figures 27 and 28 that the production of Cellulase (Cx) enzyme, took place only

FIGURE-27



PRODUCTION AND ACTIVITY OF CELLULASE (CX) ENZYME BY *Alternaria alternata* (Fries.),
Keissler. IN VIVO..

FIGURE-28



PRODUCTION AND ACTIVITY OF CELLULASE (CX) ENZYME BY *Alternaria alternata* (Fries.), Keissler.
IN VIVO..

in the diseased leaves inoculated with *Alternaria alternata* (Fries.), Keissler, in comparison to that produced in healthy leaves. The maximum loss of viscosity 46.50 per cent, was recorded after 150 minutes of standing in the diseased leaves. A gradual increase takes place pertaining to the time interval. No celluolytic activity was observed in healthy host tissues.

2. POLYGALACTURONASE (PG) :

The production of activity of Polygalacturonase (PG) enzymes by the pathogen, *Alternaria alternata* (Fries.), Keissler, was also determined by Viscometric method and the results are given in Table XXIII; and corresponding Figures 29 and 30.

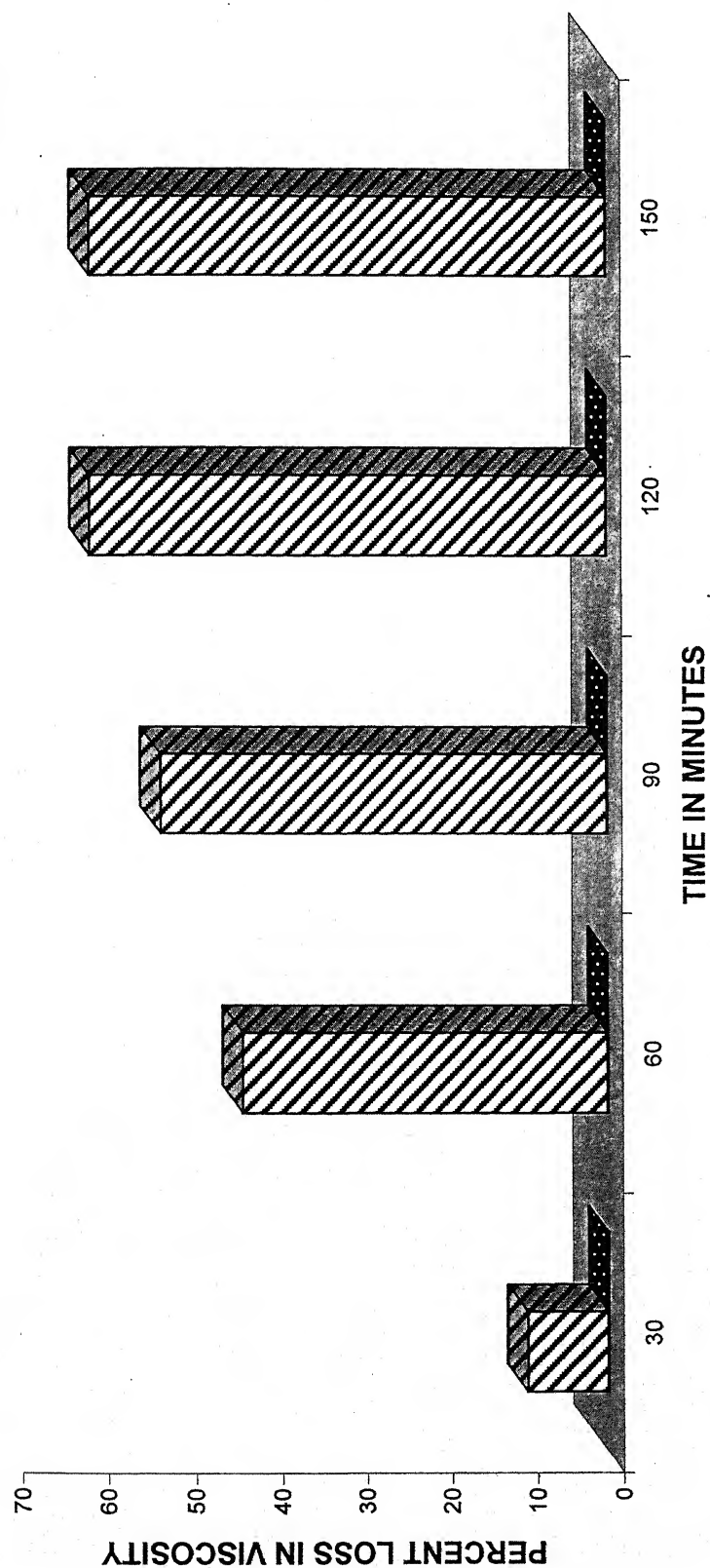
TABLE - XXIII

Production and Activity of Polygalacturonase (PG) enzyme by *Alternaria alternata* (Fries.), Keissler, in *Vivo*, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.)

Treatments	Percent Loss of Viscosity at time intervals (minutes)				
	30	60	90	120	150
Diseased	9.46	42.80	52.18	60.25	60.25
Healthy	0.0	0.0	0.0	0.0	0.0

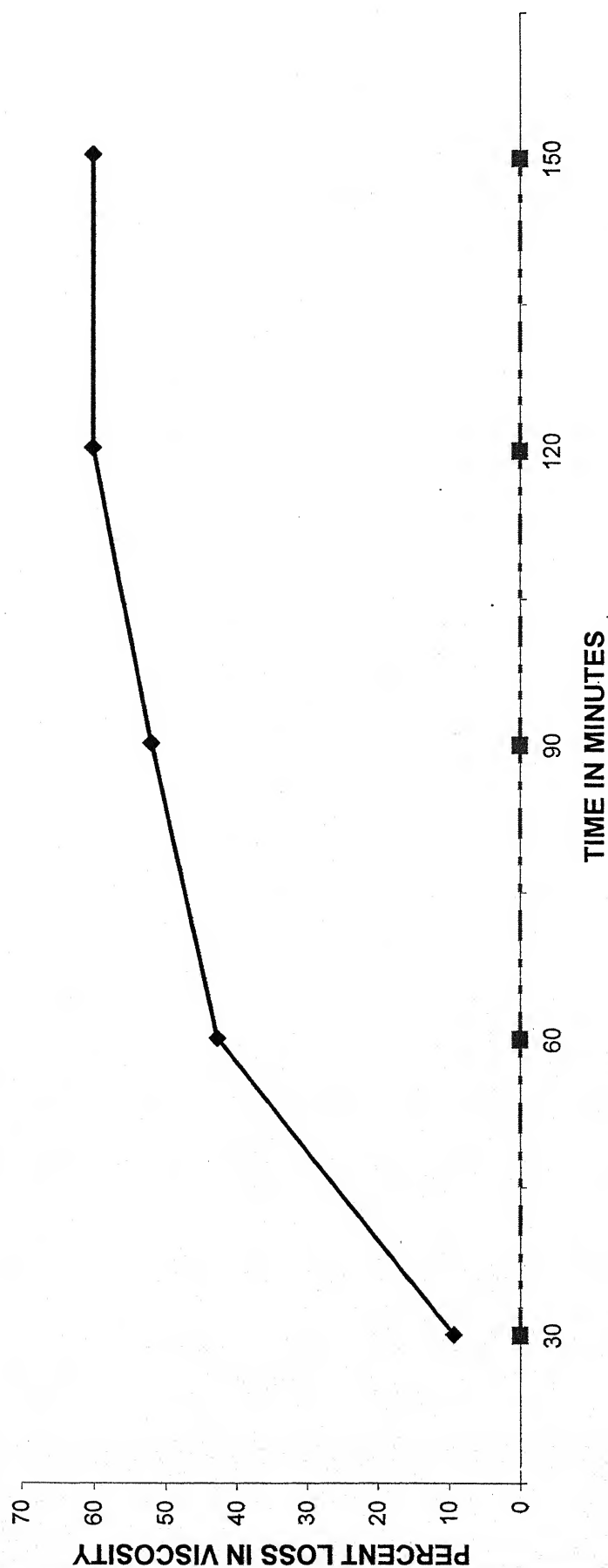
It is evident from the data presented in Table XXIII and corresponding Figures 29 and 30 that as recorded in case of Cellulase (Cx) also indicate that production of Polygalacturonase (PG) enzyme took place in the diseased leaves infected with *Alternaria alternata* (Fries.), Keissler in comparison to that produced in healthy leaves (control). The pathogen produced polygalacturonase (PG) enzyme in higher amount in the diseased leaves after 120 minutes of incubation as 60.25 per cent loss of viscosity, while there was no production of enzyme in the healthy leaves.

FIGURE-29



PRODUCTION AND ACTIVITY OF POLYGALACTURONASE (PG) ENZYME BY *Alternaria alternata* (Fries.), Keissler. IN VIVO.

FIGURE-30



PRODUCTION AND ACTIVITY OF POLYGALACTURONASE (PG) ENZYME BY *Alternaria alternata*
(Fries.), Keissler. IN VIVO..

3. POLYMETHYLGALACTURONASE (PMG)

The production and activity of Polymethylgalacturonase (PMG) enzyme by the pathogen, *Alternaria alternata* (Fries.), Keissler, was also determined by the Viscometric method and the results are given in Table XXIV and corresponding Figures 31 and 32.

TABLE - XXIV

Production and Activity of Polygalacturonase (PG) enzyme by *Alternaria alternata* (Fries.), Keissler, in *Vivo*. causing leaf spot of Dolichos bean (*Dolichos lablab*, L.)

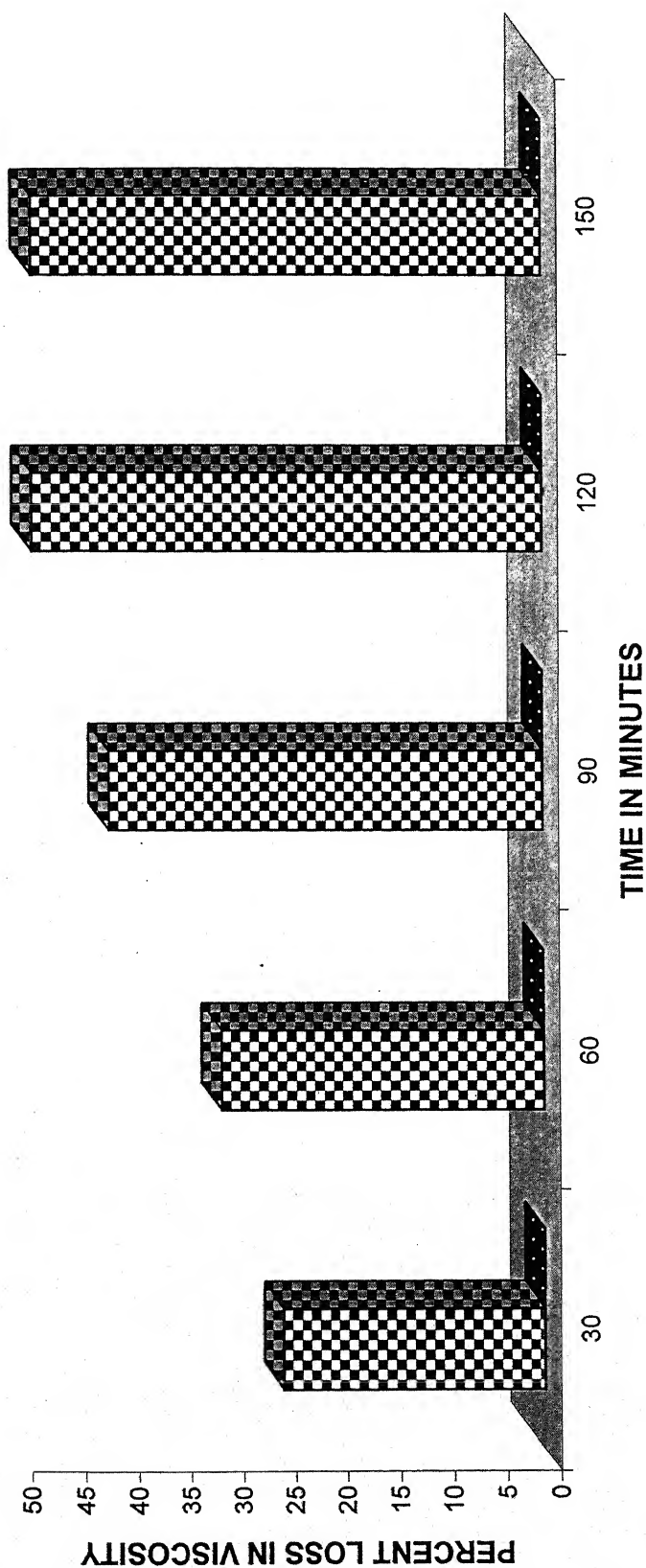
Treatments	Percent Loss of Viscosity at time intervals (minutes)				
	30	60	90	120	150
Diseased	24.70	30.60	41.20	48.30	48.30
Healthy	0.0	0.0	0.0	0.0	0.0

It is obvious from the data presented in Table XXIV and corresponding Figures 31 and 32 that production of Polymethylgalacturonase (PMG) enzyme took place in the diseased leaves infected with *Alternaria alternata* (Fries.), Keissler, in comparison to that produced in healthy leaves (control) as was in Cellulase (Cx) enzyme production. It is also evident from the fact that maximum 48.30 per cent loss of viscosity, was observed in the leaves infected with *Alternaria alternata* (Fries.), Keissler, after 120 minutes of standing. It is also clear that as the interval increases the percentage loss of viscosity (Pectinolytic activity) also increased and become constant after 120 minutes.

EFFECT OF ATMOSPHERIC TEMPERATURE, RELATIVE HUMIDITY AND RAINFALL ON DISEASE DEVELOPMENT :

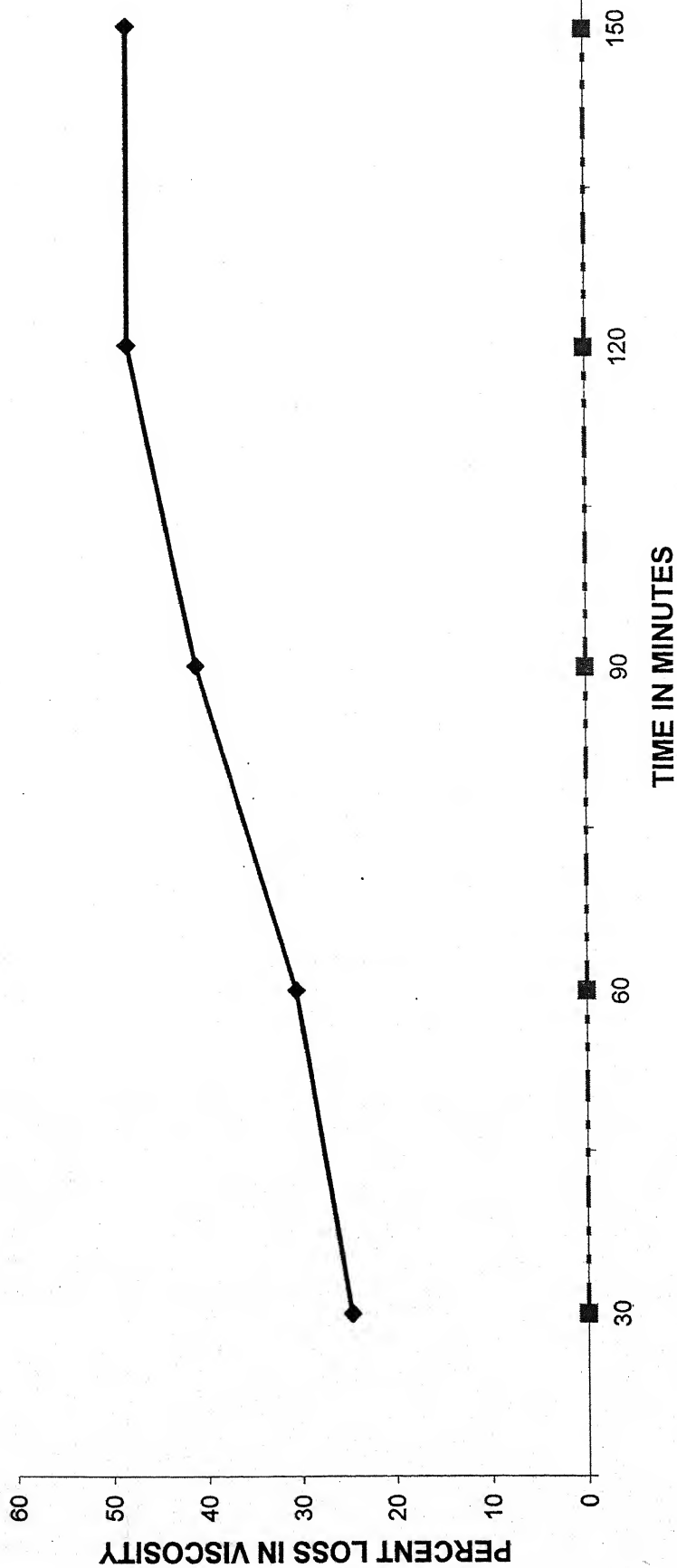
Atmospheric temperatures, relative humidities and rainfall play very

FIGURE-31



PRODUCTION AND ACTIVITY OF POLYMETHYL GALACTURONASE (PMG) EZYME BY
Alternaria alternata (Fries.), Keissler. IN VIVO..

FIGURE-32



PRODUCTION AND ACTIVITY OF POLYMETHYL GALACTURONASE (PMG) ENZYME BY *Alternaria alternata* (Fries.), Keissler. **IN VIVO.**

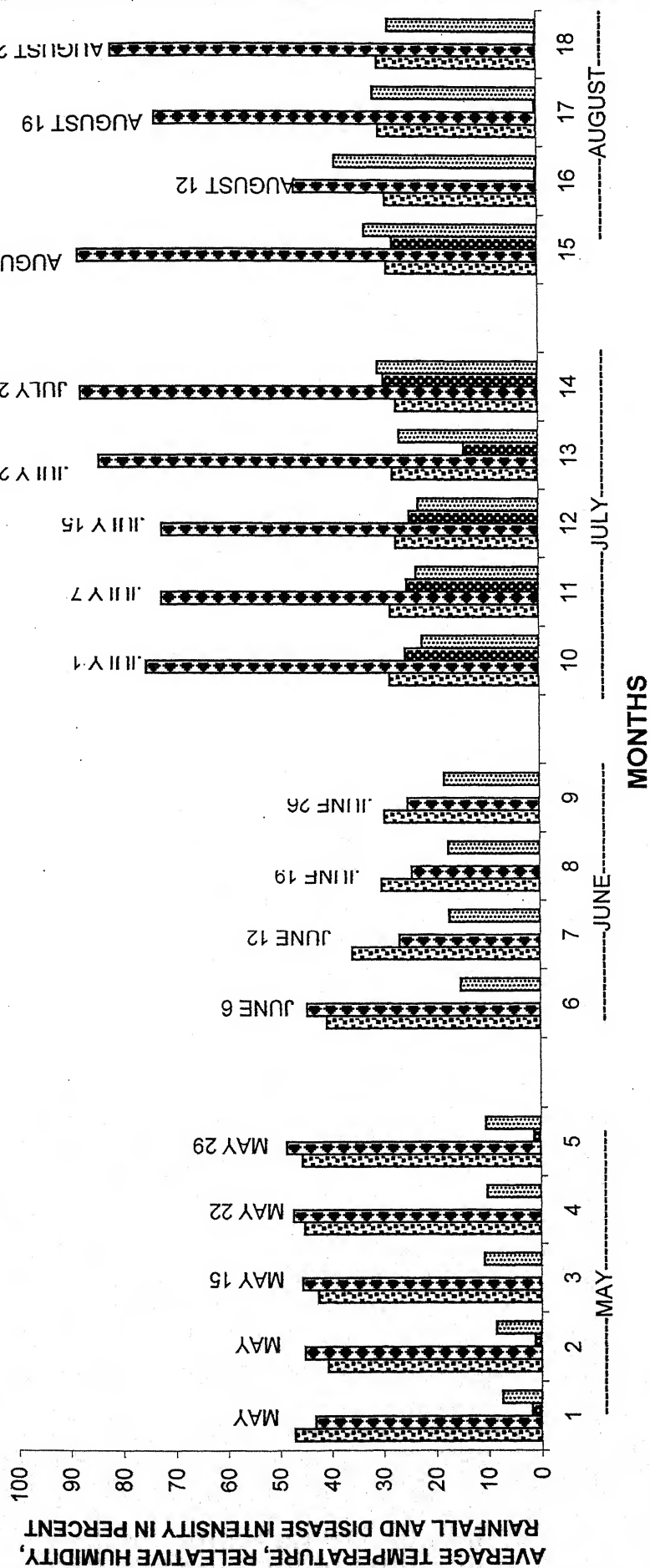
TABLE - XXV

Effect of Atmospheric temperatures, relative humidities and rainfall on leaf Spot of *Dolichos bean* (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler.

S. No.	Date	Average Temperature		Average Humidity		Total Rainfall (mm.)		Disease Intensity (0/0)	
		2001	2002	2001	2002	2001	2002	2001	2002
1.	May 1st	47.15	50.42	43.10	42.30	1.80	-	7.42	8.65
2.	May 7th	40.73	41.30	45.13	32.70	1.25	-	8.54	9.20
3.	May 15th	42.50	40.00	45.55	48.69	-	1.34	10.78	10.37
4.	May 22nd	45.15	38.20	47.34	49.15	-	2.15	10.14	12.00
5.	May 29th	45.60	39.70	48.53	46.34	1.30	-	10.40	12.60
6.	June 6th	40.83	40.08	44.57	45.83	-	-	15.03	10.30
7.	June 12th	35.75	30.20	26.67	26.37	-	-	17.09	12.00
8.	June 19th	30.10	30.85	24.25	25.48	-	-	17.25	12.40
9.	June 26th	29.52	30.97	25.03	26.82	-	-	18.04	13.47
10.	July 1st	28.38	30.14	75.30	76.45	25.40	24.52	22.18	20.60
11.	July 7th	28.25	29.23	72.42	74.87	25.10	24.20	23.29	22.70
12.	July 15th	27.19	26.18	72.29	73.57	24.57	23.60	22.80	23.10
13.	July 22	27.75	26.19	84.20	93.0	14.13	27.35	26.44	23.54
14.	July 29	27.10	28.20	87.60	80.27	29.50	21.48	30.64	25.71
15.	August 5	28.83	27.15	88.10	84.13	27.69	19.51	33.05	33.48
16.	August 12	29.05	28.70	86.70	84.25	0.29	11.48	38.74	35.57

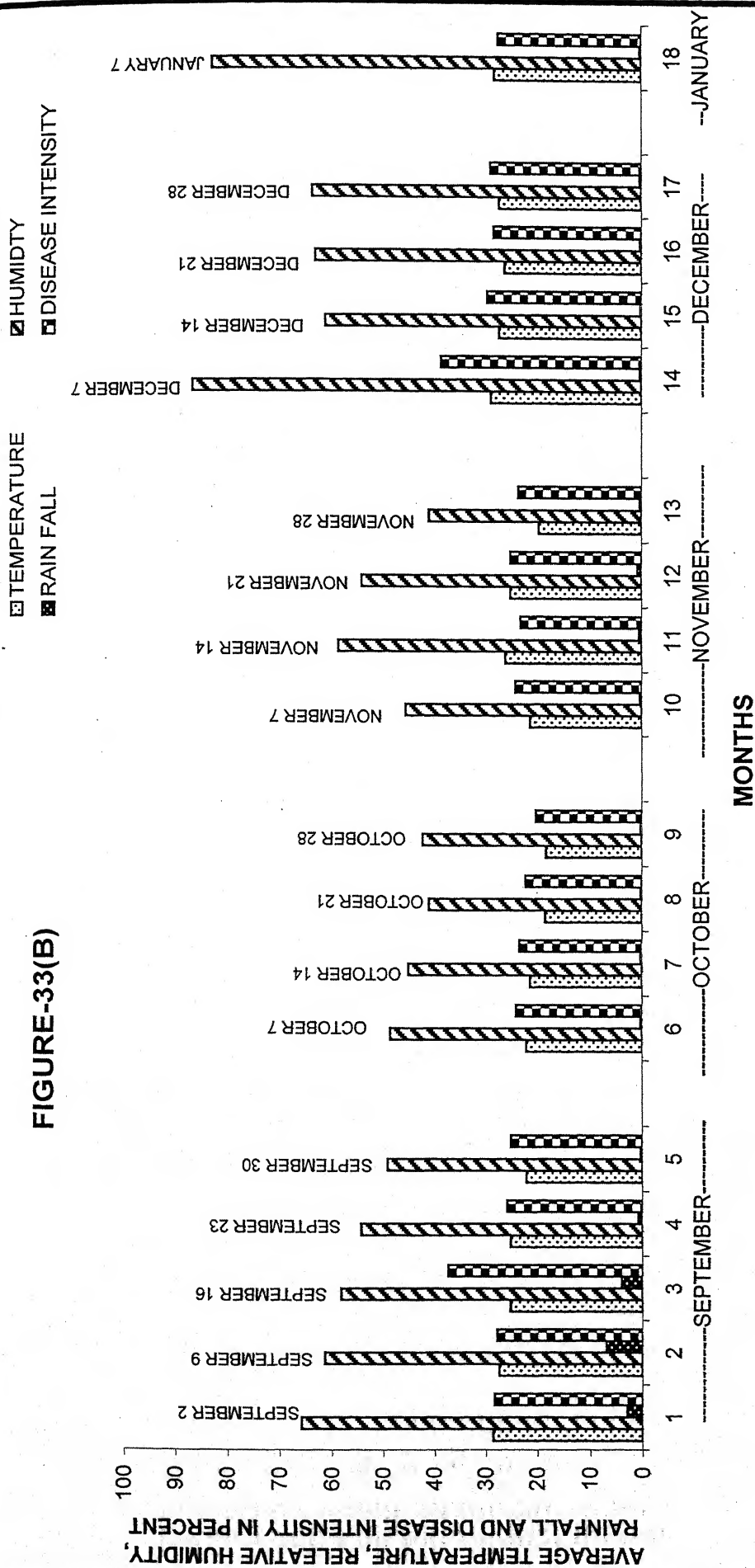
17.	August 19	30.25	28.53	83.47	79.44	0.34	7.86	31.30	34.18
18.	August 26	30.48	29.31	81.54	73.39	0.08	6.28	28.35	32.44
19.	September 2	28.60	28.09	65.83	71.47	3.00	4.97	28.35	28.84
20.	September 9	27.42	27.34	61.35	60.24	7.00	3.25	27.84	25.39
21.	September 16	25.29	24.26	58.14	56.09	4.00	2.30	27.33	25.20
22.	September 23	25.22	24.10	54.28	51.29	0.86	0.59	25.87	22.37
23.	September 30	22.14	21.24	49.17	47.82	0.33	0.27	25.19	21.39
24.	October 7	22.18	21.08	48.65	45.19	0.32	0.12	24.19	20.44
25.	October 14	21.45	19.54	45.14	43.44	0.25	0.80	23.55	19.64
26.	October 21	18.50	17.35	41.08	39.24	0.24	0.82	22.30	17.30
27.	October 28	18.42	16.45	42.27	37.58	0.21	0.0	20.30	15.48
28.	November 7	21.39	18.37	45.63	46.29	0.37	0.23	24.30	22.70
29.	November 14	26.24	17.29	58.71	60.27	0.49	0.56	23.28	20.50
30.	November 21	25.29	24.20	54.23	56.09	0.78	0.12	25.29	21.49
31.	November 28	19.70	17.39	41.12	43.54	0.27	0.15	23.75	19.74
32.	December 7	29.05	28.70	86.70	84.25	0.29	11.48	38.74	35.57
33.	December 14	27.42	27.14	61.25	60.24	0.17	6.79	29.73	26.94
34.	December 21	26.39	24.82	63.19	61.27	0.27	7.29	28.45	25.83
35.	December 28	27.42	25.39	63.78	61.94	0.29	7.39	29.12	26.37
36.	January 7	28.37	27.20	82.90	65.43	0.25	8.30	27.63	26.94

FIGURE-33(A)



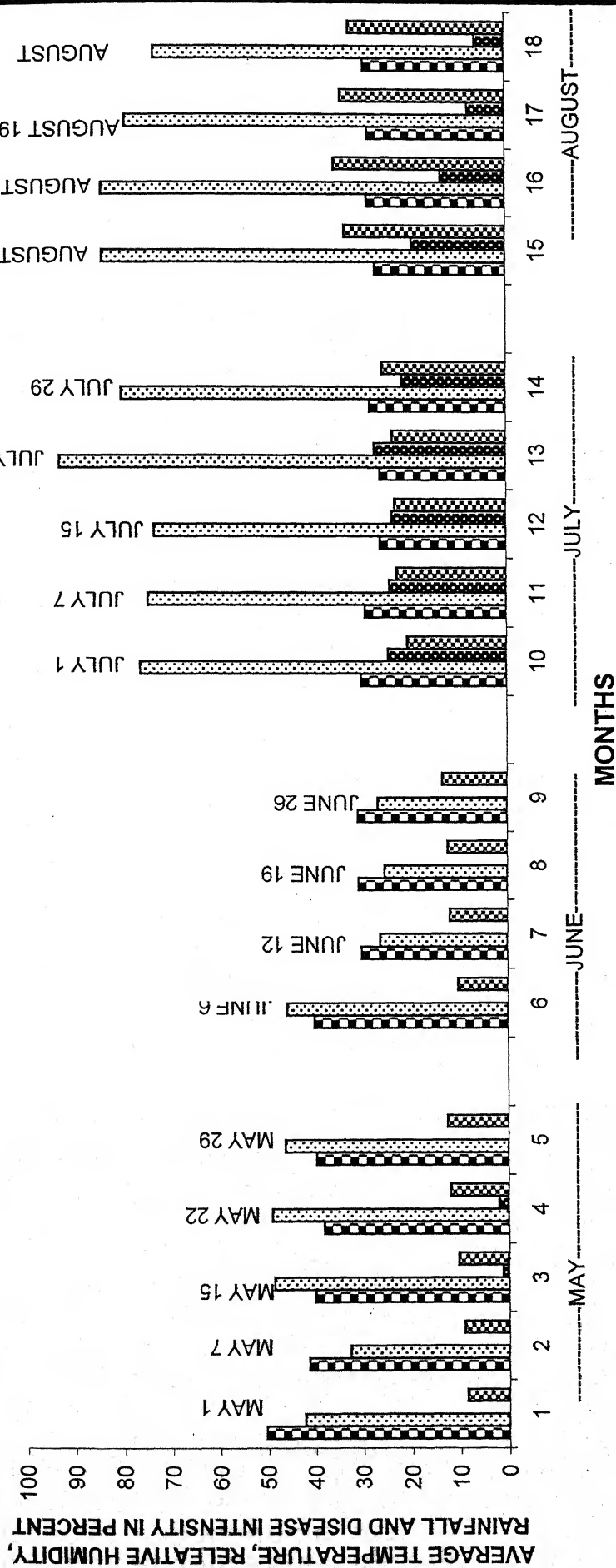
EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2001.

FIGURE-33(B)



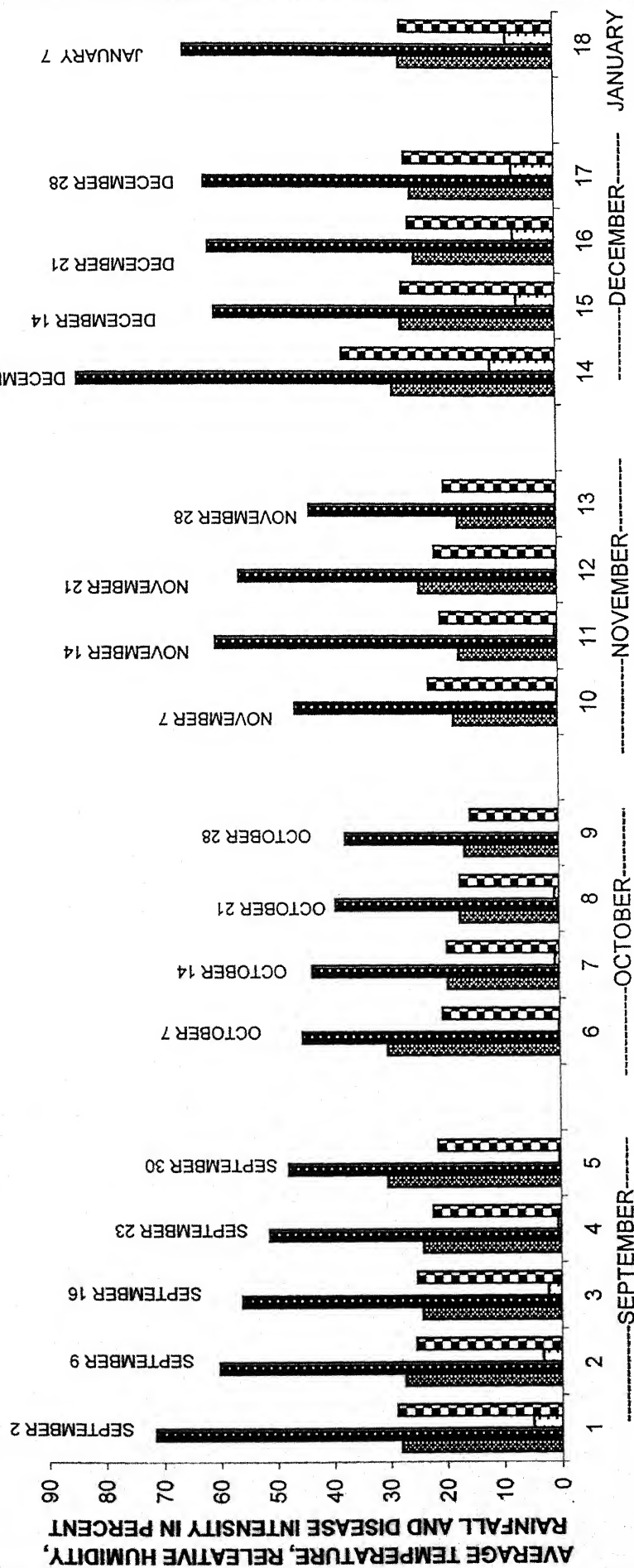
EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2001.

FIGURE-34(A)



EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2002.

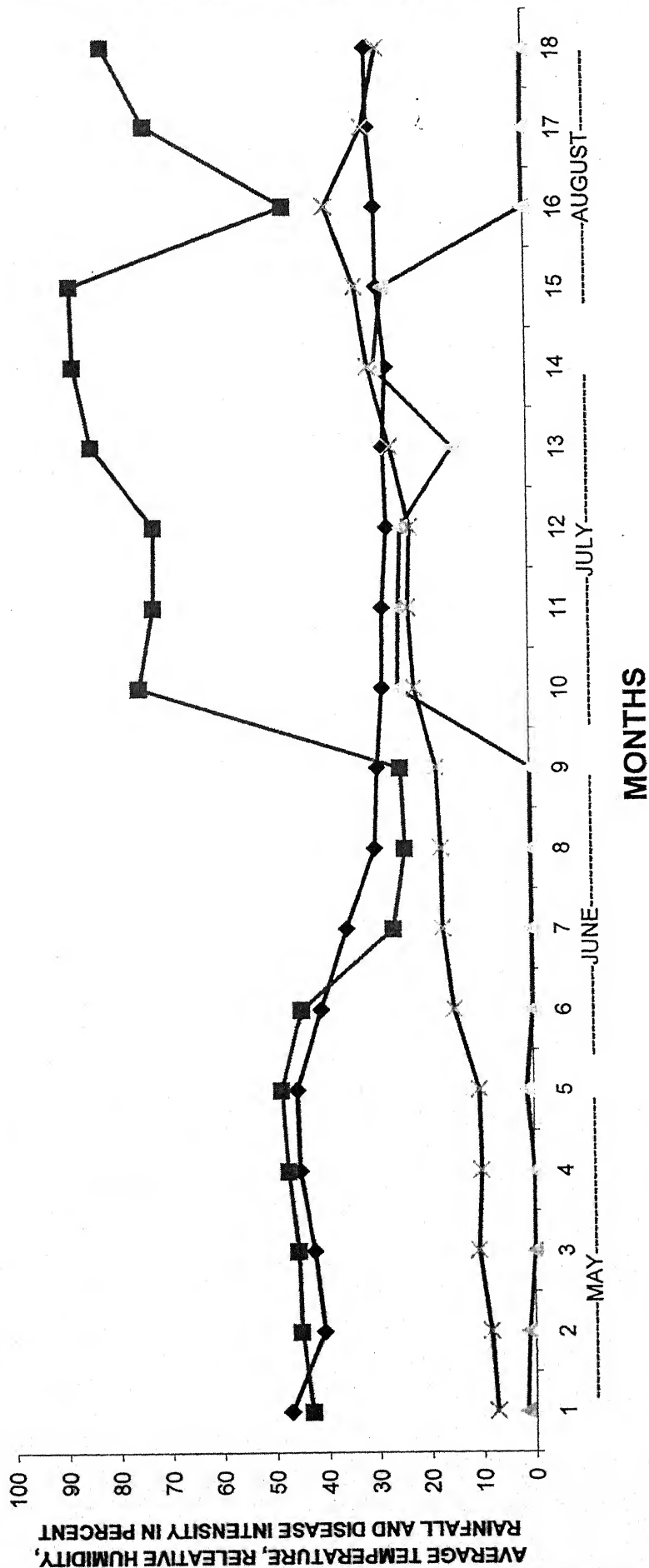
FIGURE-34 (B)



MONTHS

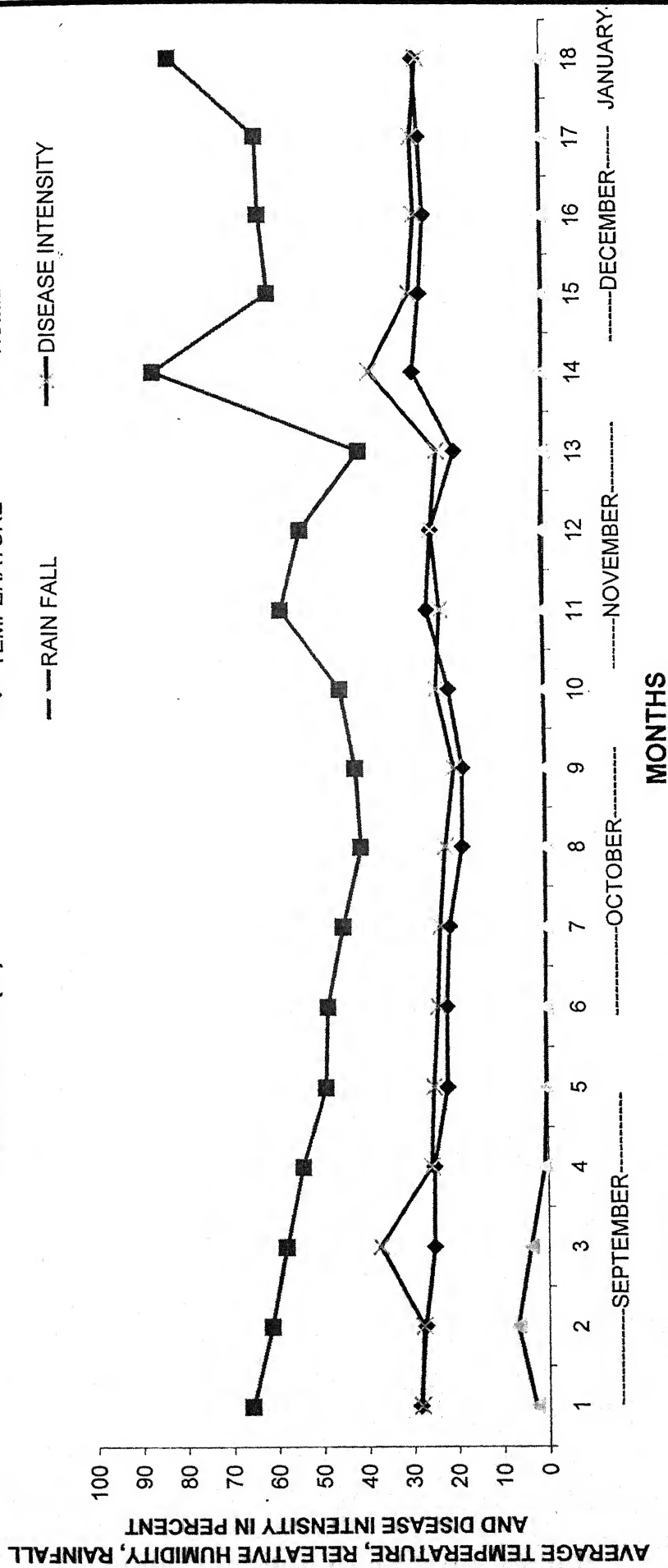
EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2002.

FIGURE-35(A)



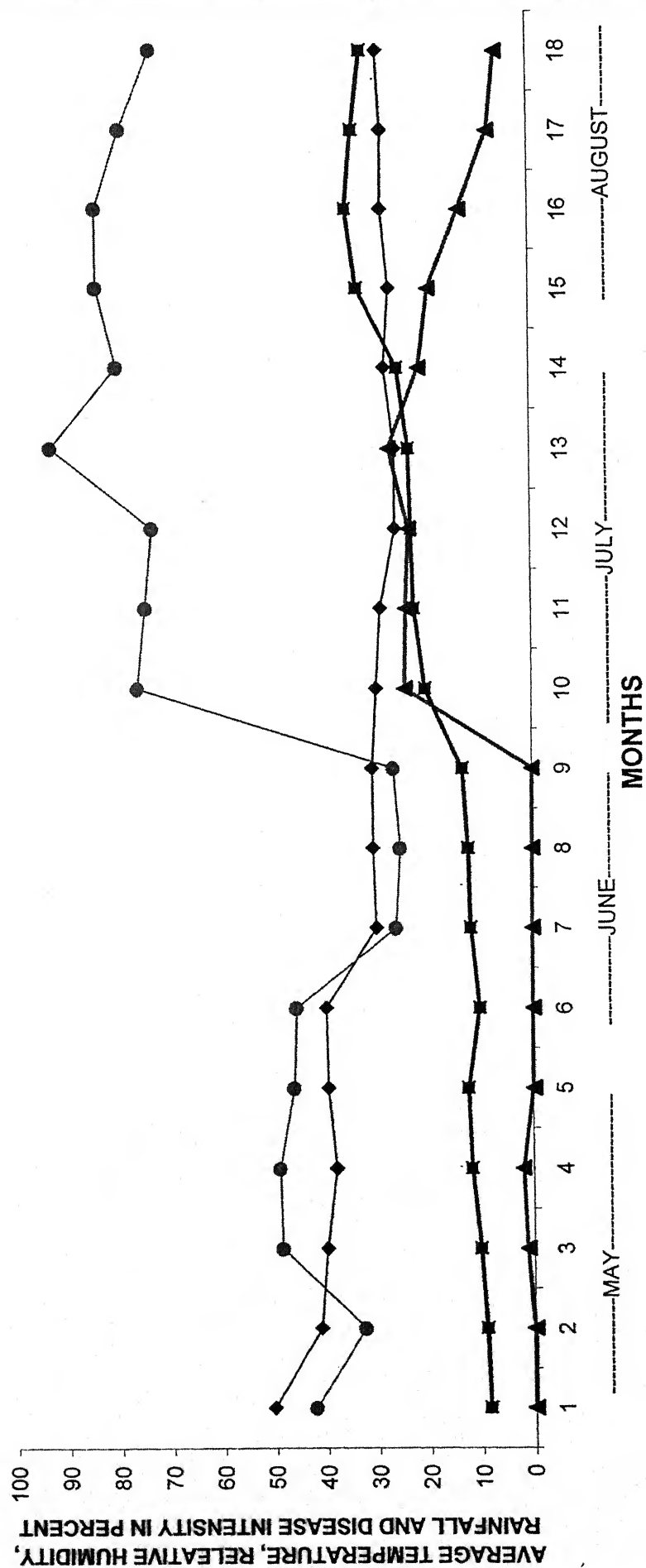
EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2001.

FIGURE-35 (B)



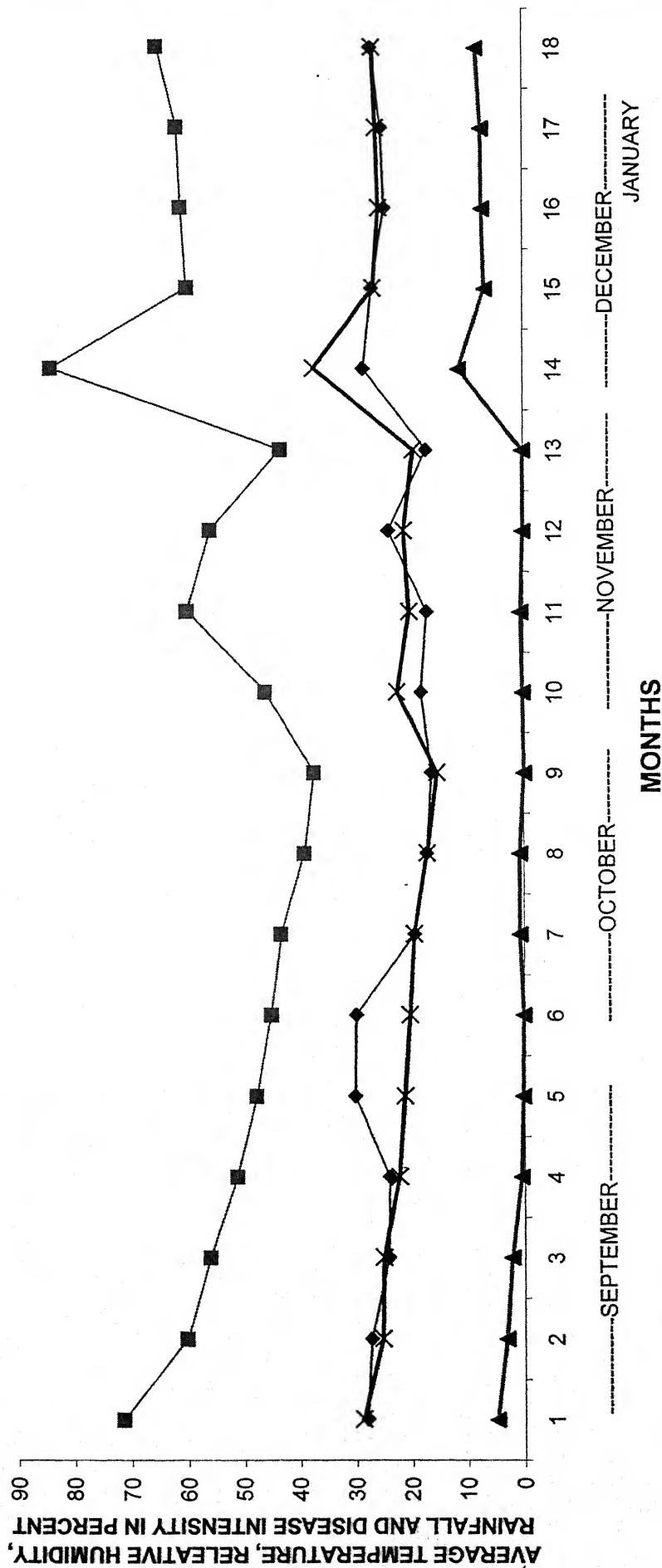
EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2001.

FIGURE-36(A)



EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2002.

FIGURE-36 (B)



EFFECT OF ATMOSPHERIC TEMPERATURES, RELATIVE HUMIDITY, RAINFALL AND DISEASE INTENSITY ON LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler. IN THE YEAR 2002.

important role in disease development, therefore it was felt desirable to study the role of these factors in the epidemiology of leaf spot of Dolichos bean caused by *Alternaria alternata* (Fries.), Keissler, by the method described under "Material and Method." The observations on disease development, were recorded at seven days interval after the appearance of the disease on the crop grown in the field. The prevailing atmospheric temperatures, relative humidity and rainfall, were also noted during the crop period and were co-related with the disease development. The individual effect of weather parameters on the disease development was analysed and results are summarised in Table XXV and Figures 33 (a and b), 34 (a and b), 35 (a and b) and 36 (a and b).

The data summarised in Table XXV and Figures 33 (a and b), 34 (a and b), 35 (a and b) and 36 (a and b) revealed that environmental factors viz., temperatures and relative humidities play an important role in disease severity. A good co-relation was recorded and established between atmospheric temperature, relative humidity and disease intensity, whereas rainfall did not affect the disease development, because it was abnormal and erratic in the years 2001 and 2002. The disease exhibited firstly its appearance in 1st week of May in both the years 2001 and 2002 and increased gradually. The maximum disease development 38.74 per cent and 35.57 per cent was recorded in the second week of August both the years 2001 and 2002 respectively, when the average temperature was 29.05°C and 28.70°C and relative humidity 86.70 and 84.25 per cent respectively. The minimum disease intensity was observed in the month of May followed by third week of October, when both the atmospheric temperature and relative humidity were unfavourable.

Thus in general, it may be concluded that disease intensity decreased with the increase in temperature, whereas it increased with the increase in relative

humidity in both the years 2001 and 2002.

NUTRITIONAL STUDIES -

In order to ascertain the effect of various sources of Carbon and Nitrogen on the growth and sporulation of the pathogen, it was grown on the above mentioned different nutritional sources on the basal medium at pH 6.50 after incubation at temperature of $25\pm 1^{\circ}\text{C}$.

GROWTH AND SPORULATION of *Alternaria alternata* (Fries.), Keissler ON DIFFERENT CARBON SOURCES IN CULTURE -

It is fact that carbon plays a significant role in nutrition of fungi. The effect of thirteen different carbon compounds was, therefore, studied in the present investigation described under, "Material and Method" for finding out the best carbon source for its growth. The average mycelial dry weight and sporulation of the pathogen, were recorded and the datas are presented in Table XXVI and Figures 37 and 38.

The results presented in Table XXVI and Figures 37 and 38, indicate that among the carbon sources tested, sucrose supported the best growth of the pathogen and was found significantly superior to all other carbon sources. Good growth of the pathogen was obtained on Galactose, Maltose, Raffinose, Dextrose, Mannose, Fructose and Xylose. Out of which the mycelial dry weight of Galactose, Maltose, Raffinose and Dextrose did not differ significantly, whereas Mannose, Fructose and Xylose differed significantly amongst themselves. The growth was fair on Sorbitol, Lactose and Mannitol amongst which the mycelial dry weight of Sorbitol and Lactose, were at par, whereas Dextrin and Rhamnose supported the poor growth of the pathogen. The poorest growth was recorded in Control, which was devoid of any carbon source.

It is also evident from the results that Sucrose, Maltose and Raffinose supported the excellent sporulation of the pathogen. Good sporulation was recorded on Galactose, Dextrose, Mannose, Fructose, Xylose and Mannitol and fair on Sorbitol.

TABLE XXVI

Average fungal dry weight and sporulation of *Alternaria alternata* (Fries.), Keissler, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.) at $25\pm 1^{\circ}\text{C}$.

S. No.	Carbon source	Average mycelial dry weight (mg.)	Sporulation
1.	Sucrose	690.10	++++
2.	Galactose	651.30	+++
3.	Maltose	637.00	++++
4.	Raffinose	607.10	++++
5.	Dextrose	591.00	+++
6.	Mannose	555.25	+++
7.	Fructose	525.10	+++
8.	Xylose	492.66	+++
9.	Sorbitol	463.00	+
10.	Lactose	450.21	+
11.	Mannitol	409.33	+++
12.	Dextrin	360.00	+
13.	Rhamnose	312.65	+
14.	Control	90.40	—

(-) = Denotes Absence of sporulation.

(+) = Denotes Poor sporulation.

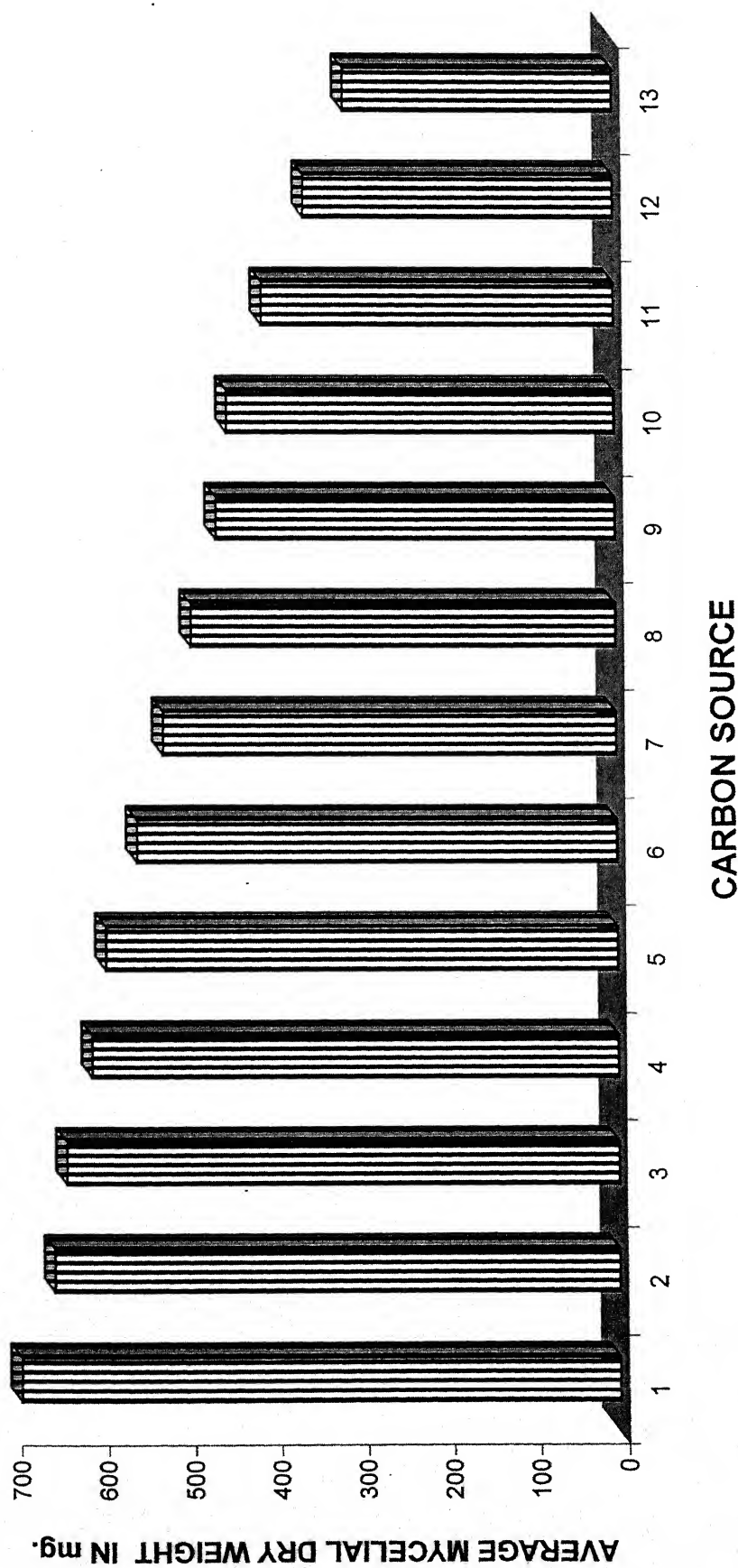
(++) = Denotes Fair sporulation.

(+++)= Denotes Good sporulation.

(++++)= Denotes Excellent sporulation.

Effect of various Carbon sources on Lactose and Dextrin, whereas poor sporulation resulted on Rhamnose. No sporulation, was recorded on the medium

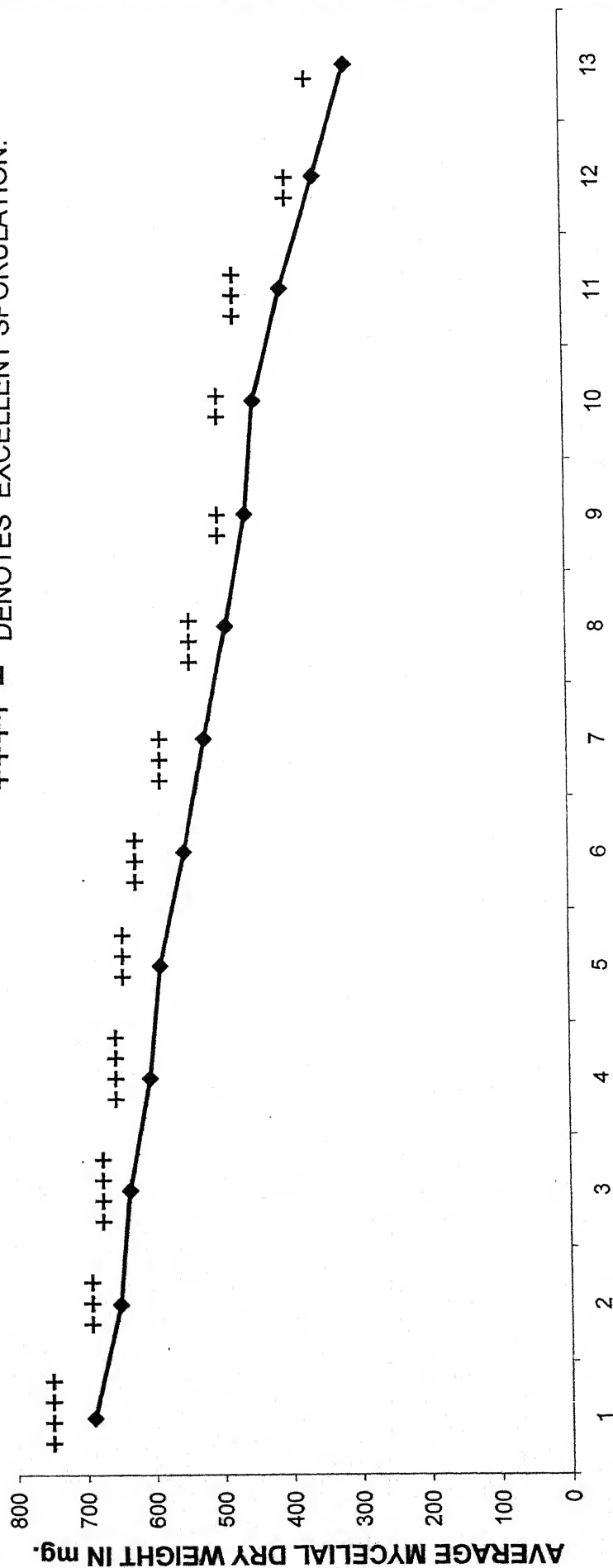
FIGURE-37



EFFECT OF DIFFERENT CARBON SOURCES ON GROWTH AND SPORULATION OF *Alternaria alternata*
(Fries.), Keissler..

FIGURE-38

- + = DENOTES POOR SPORULATION.
- ++ = DENOTES FAIR SPORULATION.
- +++ = DENOTES GOOD SPORULATION.
- ++++ = DENOTES EXCELLENT SPORULATION.



CARBON SOURCES

EFFECT OF DIFFERENT CARBON SOURCES ON GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler. .

devoid of any carbon source, which served as control.

GROWTH AND SPORULATION OF *Alternaria alternata* (Fries.), Keissler ON DIFFERENT NITROGEN SOURCES IN CULTURE -

Nitrogen is one of the most essential element utilized by the fungi for functional as well as structure purposes. The nitrogen compounds, have different nutritive values for different fungi and the later react differently to the same nitrogen source. With this aim the great selectivity of nitrogen compounds by microbes in general and fungi in particular the pathogen, was grown on twelve different organic and inorganic nitrogen sources to study its preferential requirement. Standard procedures as detailed under "Material and Method", were followed for this investigation and the data on average mycelial dry weight and sporulation are summarised in Table XXVII and Figures 39 and 40.

TABLE - XXVII

Effect of various nitrogen sources on the average fungal dry weight and sporulation of *Alternaria alternata* (Fries.), Keissler, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.) at $25\pm 1^{\circ}\text{C}$.

S. No.	Nitrogen Sources	Average mycelialdry weight (mg.)	Sporulation
1.	Ammonium acetate	365.00	+
2.	Ammonium carbonate	242.00	+
3.	Ammonium chloride	474.00	+
4.	Ammonium oxalate	418.30	+
5.	Ammonium sulphate	304.66	+
6.	Ammonium nitrate	501.20	+++
7.	Calcium nitrate	571.00	+++
8.	Peptone	845.00	++++
9.	Potassium nitrate	640.00	+++
10.	Sodium nitrate	734.10	++++

11.	Thiourea	144.21	—
12.	Urea	171.33	+
13.	Control	119.00	—

(-) = Denotes Absence of sporulation.

(+) = Denotes Poor sporulation.

(++) = Denotes Fair sporulation.

(+++)= Denotes Good sporulation.

(++++)= Denotes Excellent sporulation.

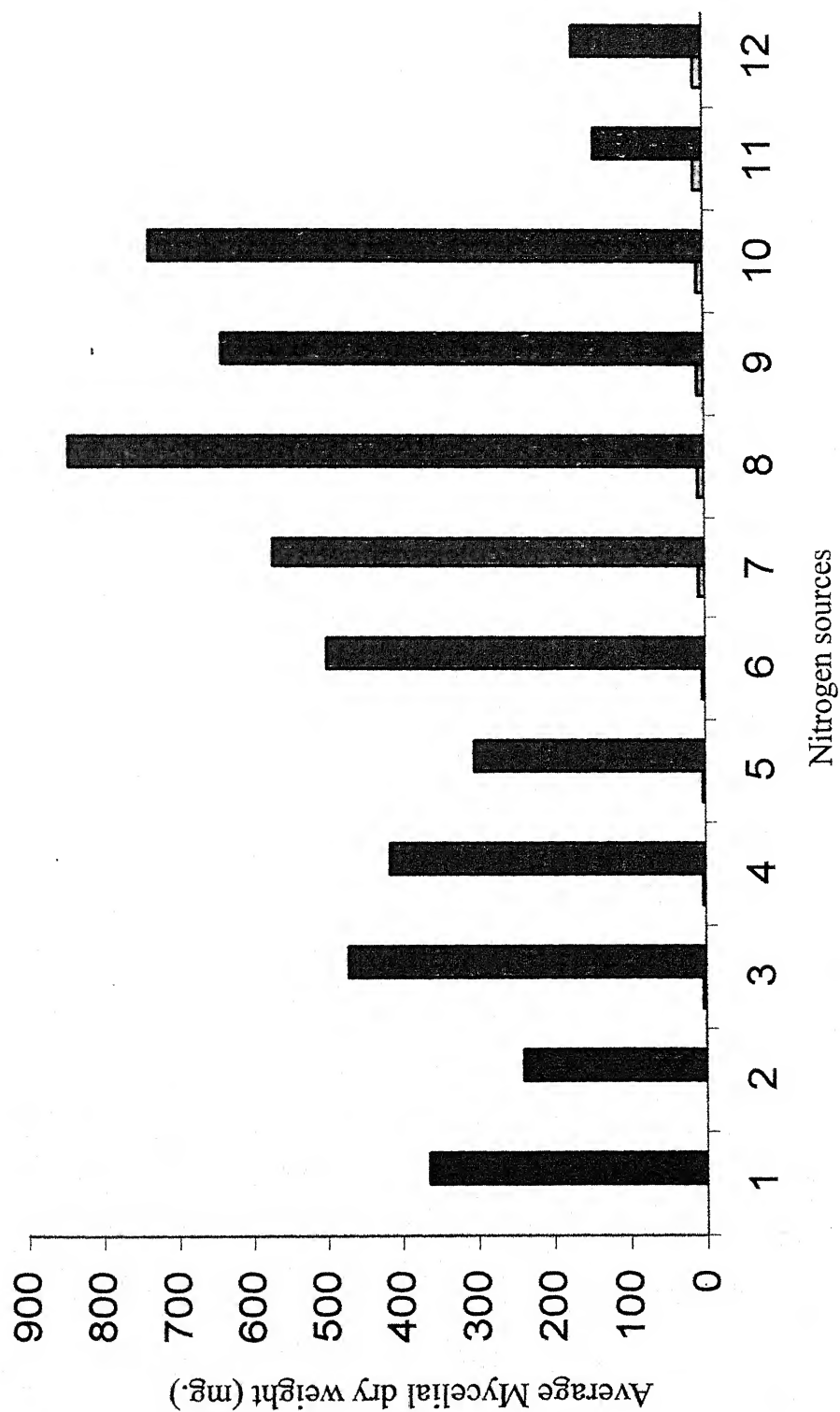
It is evident from the results expressed in Table XXVII and Figures 39 and 40 that Peptone supported the best growth of pathogen and was significantly superior to the rest of the treatments. Good growth of the fungus, was recorded on ammonium chloride, ammonium nitrate, calcium nitrate, potassium nitrate and sodium nitrate. Out of which ammonium nitrate and ammonium chloride did not differ significantly. Fair growth was obtained on ammonium acetate, ammonium oxalate and ammonium sulphate. Although ammonium carbonate, thiourea and urea exhibited poor growth, but urea and thiourea exhibited similar response. The lowest growth was obtained in control, which was devoid of any nitrogen source.

Excellent sporulation was recorded in the medium supplemented with peptone and sodium nitrate as nitrogen source, whereas ammonium nitrate, calcium nitrate and potassium nitrate exhibited good sporulation. Sporulation was fair in the medium substituted with ammonium acetate, ammonium chloride and ammonium oxalate as nitrogen source, whereas poor sporulation was observed in ammonium carbonate, ammonium sulphate and Urea. No sporulation was recorded in thiourea and control.

EFFECT OF DIFFERENT DOSES OF NITROGEN, PHOSPHORUS AND POTASH ON THE SEVERITY OF DISEASE -

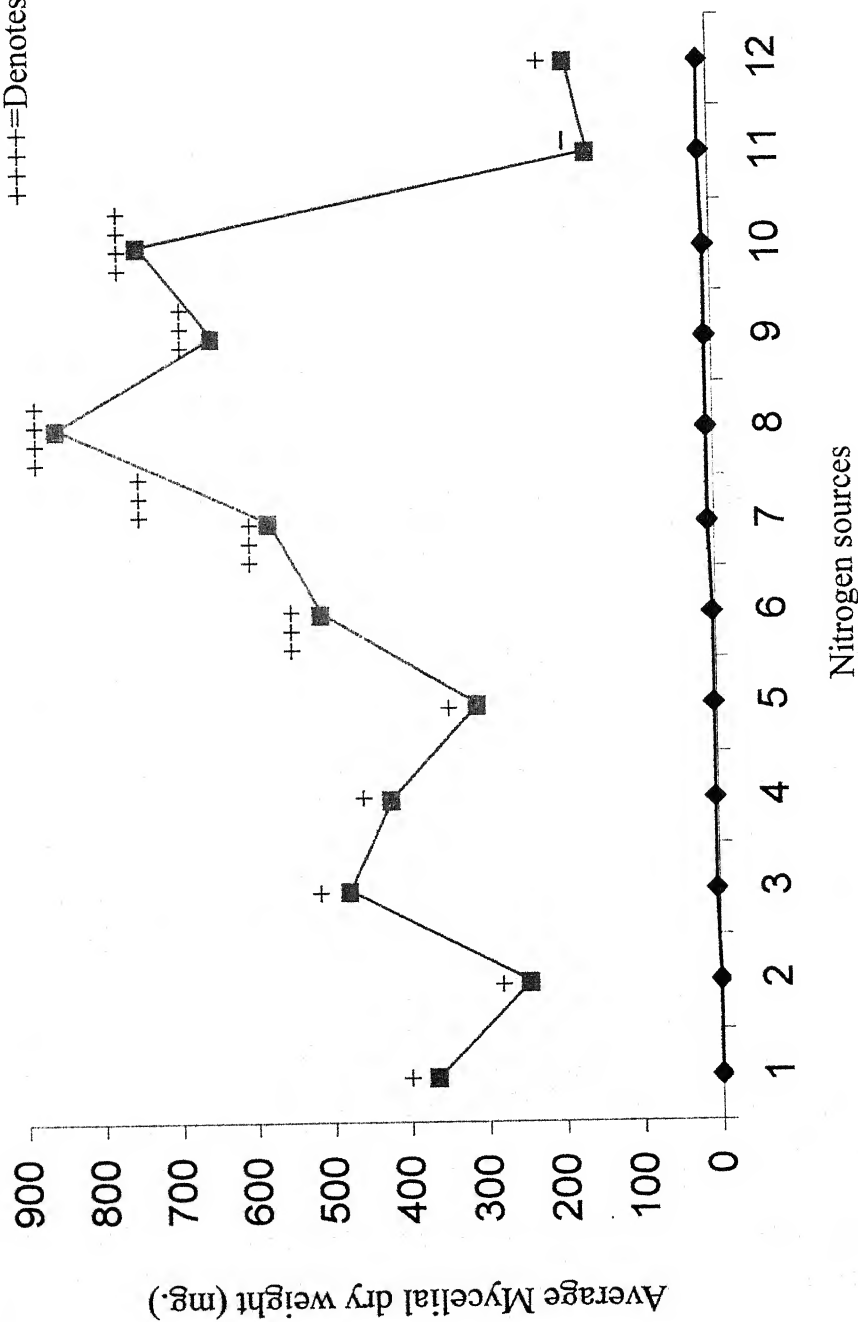
In order to determine the effect of nitrogen, phosphorus and potash on the

Figure-39



Effect of various nitrogen sources on the average fungal dry weight and sporulation of *Alternaria alternata* (Fries.), Keissler, causing leaf spot of Dolichos bean (*Dolichos lablab*, L.) at $25 \pm 1^\circ\text{C}$

Figure-40



- Denotes absence of sporulation
 += Denotes poor sporulation
 ++= Denotes fair sporulation
 +++= Denotes good sporulation
 ++++= Denotes excellent sporulation

Effect of various nitrogen sources on the average fungal dry weight and sporulation of *Alternaria alternata* (Fries.), Keissler, causing leaf spot of *Dolichos lablab*, L.) at $25 \pm 1^\circ\text{C}$

severity of disease, experiments with different combination of fertilizers were conducted in the field by sowing highly susceptible variety "Kalyanpur Type - 1" during Kharif season of 2001 and 2002. Disease intensity was recorded at harvest time on the basis of percentage leaf area affected. Data and results are summarised in Table XXVIII and XXIX.

TABLE - XXVIII

Effect of NPK on the intensity of Leaf spot of Dolichos bean (*Dolichos lablab*, L.) during Kharif season of the year 2001.

Treat-ments	N0	N60	N120	Mean	K0	K40	Mean
P ₀	24.02 (29.35)	26.74 (31.14)	31.59 (34.20)	27.39 (31.56)	32.88 (34.94)	22.22 (28.13)	27.39 (31.56)
P ₆₀	21.98 (27.96)	23.96 (29.31)	29.85 (33.12)	25.19 (30.13)	30.11 (33.28)	20.59 (26.98)	25.19 (30.13)
Mean	23.00 (28.66)	25.35 (30.23)	30.72 (33.66)	— —	34.48 (34.13)	21.40 (27.56)	— —
K ₀	27.37 (31.55)	30.38 (33.45)	36.89 (37.40)	31.48 34.13	— —	— —	— —
K ₄₀	18.88 (25.76)	20.61 (27.00)	24.86 (29.91)	21.40 (27.56)	— —	— —	— —
Mean	23.00 (28.66)	25.34 (30.23)	30.72 (33.66)	— —	— —	— —	— —

EFFECT OF NITROGEN (N) :-

The results presented in Tables XXVIII and XXIX indicate that the effect of nitrogen levels, was highly significant as regards to disease development in both the years. It is also obvious that disease intensity increased with the increase in the nitrogen doses. Maximum disease intensity was recorded at the application of 120 Kg. nitrogen per hectare, where it was lowest in control, where nitrogen was not added to the soil in both the years. It may also be observed from Table that disease intensity increased three times more over control, while nitrogen doses increased only two times.

TABLE XXIX

Effect of NPK on the intensity of Leaf spot of *Dolichos bean* (*Dolichos lablab*, L.) during Kharif season of the year 2002.

Treat-ments	N0	N60	N120	Mean	K0	K40	Mean
P ₀	25.15 (10.10)	27.20 (31.44)	32.65 (34.85)	28.28 (33.13)	33.98 (35.66)	22.91 (28.60)	28.28 (32.13)
P ₆₀	23.01 (28.67)	24.96 (29.98)	30.75 (33.68)	26.18 (30.78)	31.10 (33.90)	21.53 (27.65)	26.18 (30.78)
Mean	24.06	26.08	31.69	—	32.53	22.22	—
K ₀	28.49 (32.26)	31.38 (34.07)	37.90 (38.00)	32.53 (34.78)	— —	— —	— —
K ₄₀	19.90 (26.50)	21.10 (27.35)	25.80 (30.53)	22.22 (28.13)	— —	— —	— —
Mean	24.86 (29.38)	26.08 (30.71)	31.69 (34.26)	— —	— —	— —	— —

EFFECT OF PHOSPHORUS -

Application of phosphorus to the soil decreased the disease intensity (Tables XXVIII and XXIX) over control in both the years. The decrease in disease due to phosphorus application was 4.53 and 4.20 per cent over control in Kharif crop season during 2001 and 2002 respectively.

EFFECT OF POTASH (K) -

Disease intensity was significantly reduced, when potash was applied to soil during Kharif season crop, 2001 and 2002. However it was 19.25 per cent and 19.12 per cent over control in both the years respectively.

EFFECT OF NXP -

The effect of the interaction of nitrogen and phosphorus, was found non-significant in both the years of experimentation.

EFFECT OF NXX -

The interaction of nitrogen and potash was found significant during, 2002, while it was non-significant in the year, 2001. At the same levels of potash (K_0 and K_{40}), nitrogen at 60.0 kg. per hectare had increased the disease intensity in comparison to control. Similarly, at the same levels of nitrogen and potash at 40.0 kg. per hectare had increased the disease.

EFFECT OF PXX -

It is obvious from the results that the effect of phosphorus and potash on disease development was found non-significant in the year, 2001, while it was significant in the year, 2002. However the combination of $P_{60} K_{40}$ (27.65) was found superior.

EFFECT OF NXPXX -

The interaction of NPK, was found significant during, both the years. The combination of N_{120}, P_0K_0 , had the highest disease intensity (38.70 per cent and 39.28 per cent) for the year 2001 and 2002 respectively.

SUSCEPTIBLE GROWTH PERIOD OF DOLICHOS BEAN (*Dolichos lablab*, L.) PLANTS IN RELATION OF DISEASE DEVELOPMENT :

Every disease has maximum disease development at a particular time of its susceptibility, which helps in suggesting prophylactic measures against the incidence of disease at right time. The present study was therefore undertaken by the procedure given under, "Material and Method" in order to find out the most susceptible age of plants. For this study surface sterilized seeds of Dolichos bean germplasm/culture "Kalyanpur type - 1", were sown in sterilized soil in 30.0 cm. earthen pots. Four plants were maintained and for each sowing date three pots were used. The sowings were done at 10 days interval so that plants attained the age from 10 days to 90 days. These plants were inoculated with the mycelial-cum-spore suspension of *Alternaria alternata* (Fries.), Keissler, when the first sown plants attained the age of 90 days. Final data on disease intensity was recorded after 15 days of inoculation and results are summarised in Table

XXX and Figures 41 and 42.

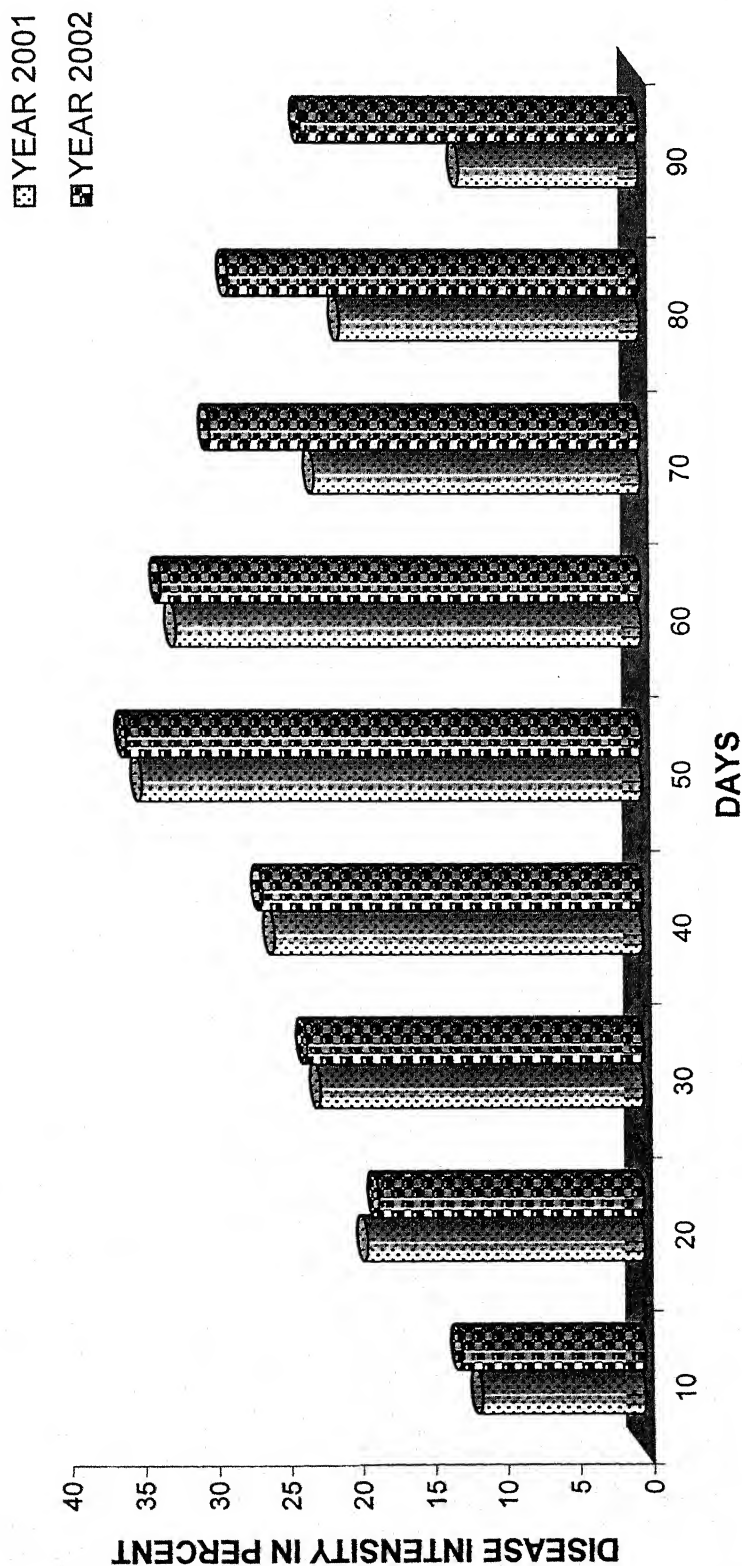
TABLE - XXX

Susceptible growth period of Dolichos bean (*Dolichos lablab*, L.) plants in relation to disease development causing leaf spot caused by *Alternaria alternata* (Fries.), Keissler.

S.No.	Age of Plant (Days)	Disease Intensity in Per cent	
		Year 2001	Year 2002
1.	10	11.23	12.65
2.	20	19.10	18.34
3.	30	22.30	23.26
4.	40	25.47	26.28
5.	50	34.53	35.60
6.	60	32.17	33.18
7.	70	22.60	29.83
8.	80	20.75	28.50
9.	90	12.47	23.42

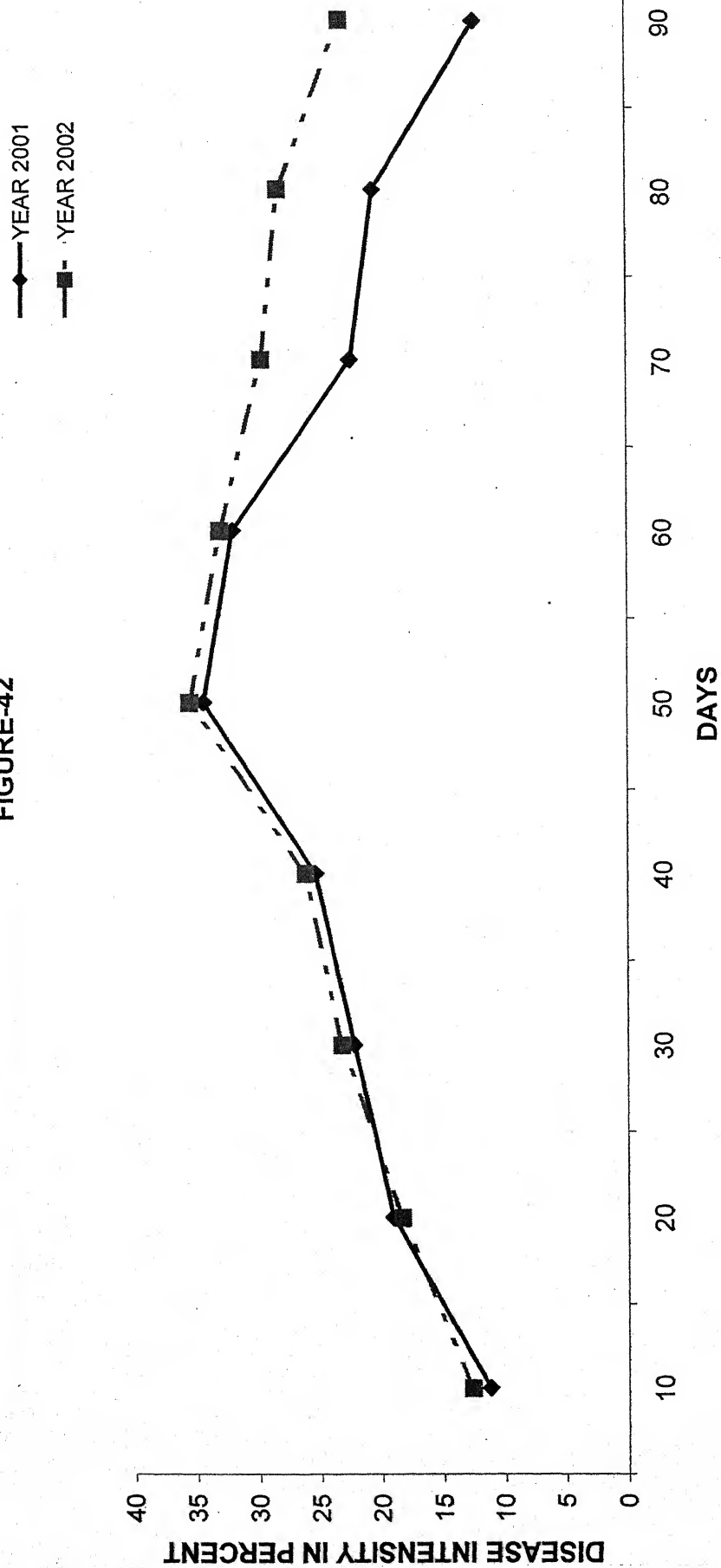
It is obvious from the Table XXX and corresponding Figures 41 and 42 that the pathogen, *Alternaria alternata* (Fries.), Keissler infected the Bean plants in all the ages ranging from 10 days to 90 days. The maximum disease intensity was observed in the plants, which attained the age of 50 days followed by 60 and 40 and 60 to 70 days old plants during both the years respectively. The minimum disease intensity was recorded from 10 days old plants. Thus it was concluded that Dolichos bean plants were susceptible to disease at the age of 40 to 60 and 50 to 70 days particularly at 50 days respectively in both the years. The susceptibility of plants towards disease decreased with the increasing age of plants and found almost minimum at the age of 90 days and onward.

FIGURE-41



SUSCEPTIBLE GROWTH PERIOD (AGE OF PLANTS) IN RELATION TO DISEASE DEVELOPMENT OF DOLICHOS BEAN (*Dolichos lablab* L.)

FIGURE-42



SUSCEPTIBLE GROWTH PERIOD (AGE OF PLANTS) IN RELATION TO DISEASE DEVELOPMENT OF DOLICHOS BEAN (*Dolichos lablab* L.)

In general the susceptibility of the plants to the disease gradually was found decreased below or above 50 days old plants.

CHANGES IN BIOCHEMICAL CONSTITUENTS OF BEAN LEAVES IN RELATION TO DISEASE DEVELOPMENT.

In order to study the biochemical parameters relating to disease resistance, susceptibility of leaf spot of Dolichos bean the healthy and diseased leaves were analysed according to the technique described under "Material and Method". The contents of wax, chlorophyll, polyphenol, sugar, nitrogen, phosphorus, potassium and sulphur, were observed and results are summarised in Table XXXI and Figures 43, 44, 45, 46 and 47.

TABLE - XXXI

Chemical constituents of healthy and diseased leaves of Dolichos bean (*Dolichos lablab*, L.) at different stages after inoculation by *Alternaria alternata* (Fries.), Keissler.

Parameters	Healthy (Uninoculated leaves)	Inoculated Leaves		
		After 40 Days	After 70 Days	Necrotic tissue after 70 days
Wax	28.70 mg./g.	24.20 mg./g.	18.40 mg./g.	4.50 mg./g.
Chlorophyll "a"	0.85 mg./g.	0.68 mg./g.	0.27 mg./g.	0.15 mg./g.
Chlorophyll "b"	1.78 mg./g.	1.34 mg./g.	0.75 mg./g.	0.33 mg./g.
Polyphenols	9.75 mg./g.	7.45 mg./g.	3.60 mg./g.	1.30 mg./g.
Reducing sugars	13.17 mg./g.	11.20 mg./g.	8.46 mg./g.	5.93 mg./g.
Non-reducing sugars	5.80 mg./g.	4.20 mg./g.	1.39 mg./g.	1.33 mg./g.
Nitrogen	3.45%	2.75%	1.55%	1.20%

Phosphorus	0.25%	0.38%	0.17%	0.15%
Potash	2.85%	2.84%	1.49%	1.10%
Sulphur	0.55%	0.52%	0.38%	0.35%

From the results summarised in Table XXXI, and Figures 43, 44, 45, 46 and 47, it is evident that after 40 and 70 days of inoculation of healthy leaves by the pathogen *Alternaria alternata*, considerable changes in the amount of different chemical constituents of diseased leaves were observed as compared to healthy leaves. It was also observed that contents of wax, chlorophyll "a" and "b", polyphenols, reducing and non-reducing sugars and nitrogen were comparatively decreased in both the categories of inoculated leaves in descending order by utilizing them its own food requirement or by destroying them through reaction. A reduction in amount of wax, "chlorophyll a" and "b", polyphenols and reducing sugars in the necrotic tissues of the leaves after 70 days of inoculation were observed but no remarkable changes, were recorded in the contents of phosphorus, potash and sulphur.

In general, it is concluded that reduction in the amount of chemical constituents of diseased leaves was directly associated with disease severity.

MODE OF SURVIVAL OF PRIMARY INFECTION AND SPREAD OF DISEASE :

To minimize the losses, knowledge of mode of survival of primary source of infection, perpetuation and spread of disease, is essential for planning the effective control measures therefore role of seeds, infested soil and disease plants debris in the survival of *Alternaria alternata* (Fries.), Keissler and primary infection of the disease was separately studied in detail by the technique described under, "Material and Method."

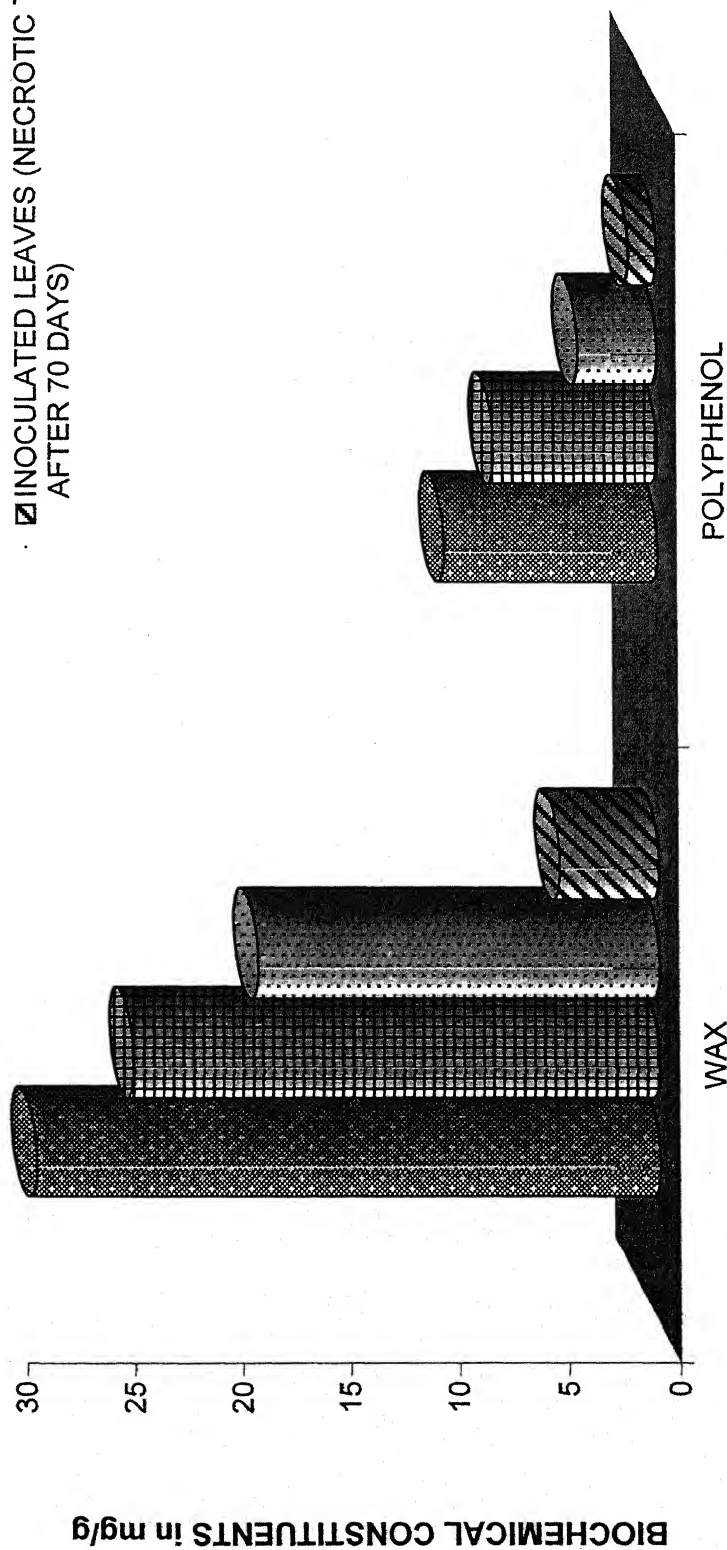
1. ROLE OF SEED IN SURVIVAL OF PLANT PATHOGEN :

(a) UNDER LABORATORY :

To determine the role of seeds in survival of pathogen, *Alternaria alternata*

FIGURE-43

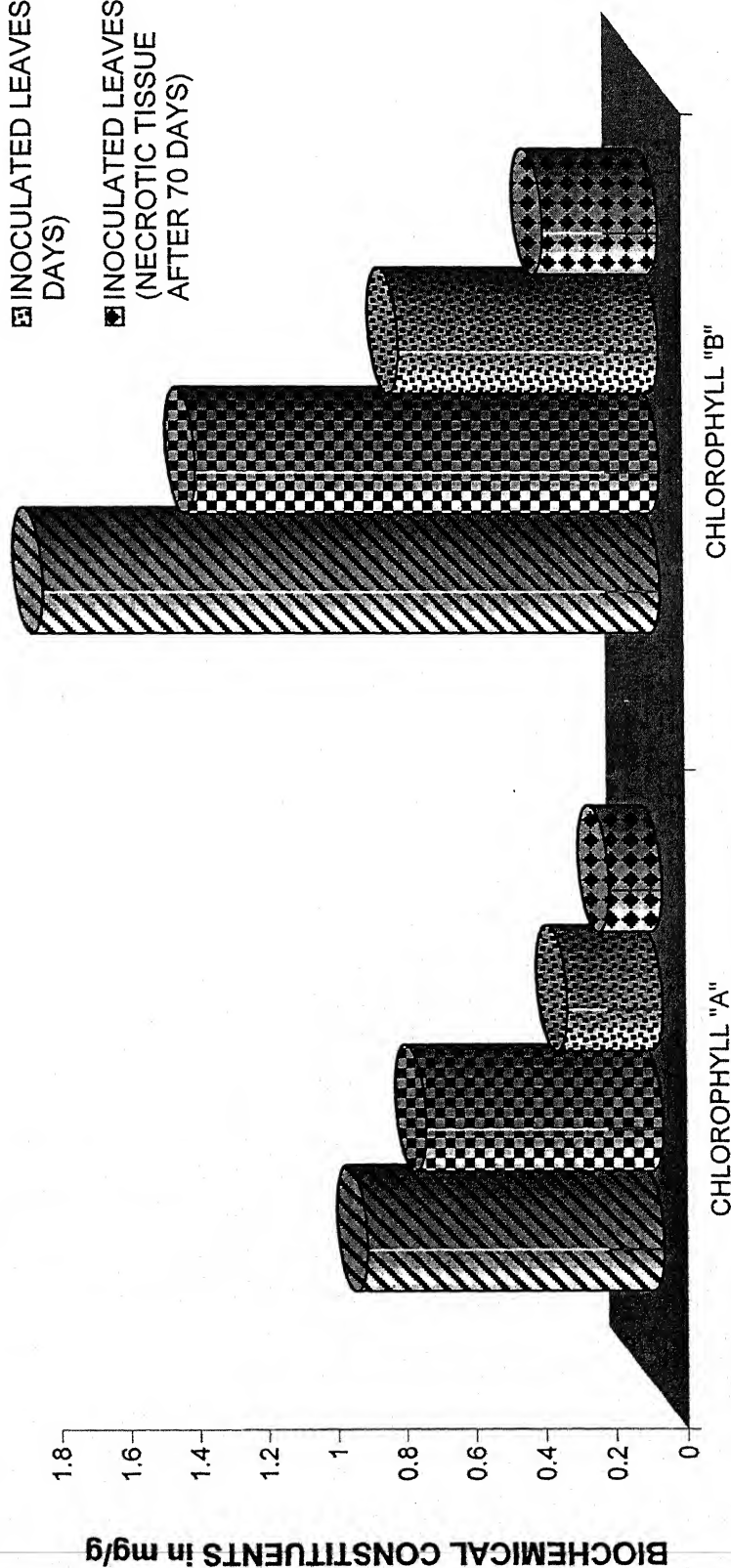
- UN-INOCULATED LEAVES
- ▨ INOCULATED LEAVES (40 DAYS)
- ▤ INOCULATED LEAVES (70 DAYS)
- ▩ INOCULATED LEAVES (NECROTIC TISSUE AFTER 70 DAYS)



WAX AND POLYPHENOL CONTENTS IN LEAVES OF DOLICHOS BEAN
(*Dolichos lablab*, L.)

FIGURE-44

- UN-INOCULATED LEAVES
- INOCULATED LEAVES (40 DAYS)
- INOCULATED LEAVES (70 DAYS)
- INOCULATED LEAVES (NECROTIC TISSUE AFTER 70 DAYS)



CHLOROPHYLL "A" AND CHLOROPHYLL "B" IN LEAVES OF DOLICHOS BEAN
(*Dolichos lablab*, L.)

FIGURE-45

- UN-INOCULATED LEAVES
- ▨ INOCULATED LEAVES (40 DAYS)
- ▤ INOCULATED LEAVES (70 DAYS)
- ▥ INOCULATED LEAVES (NECROTIC TISSUE AFTER 70 DAYS)

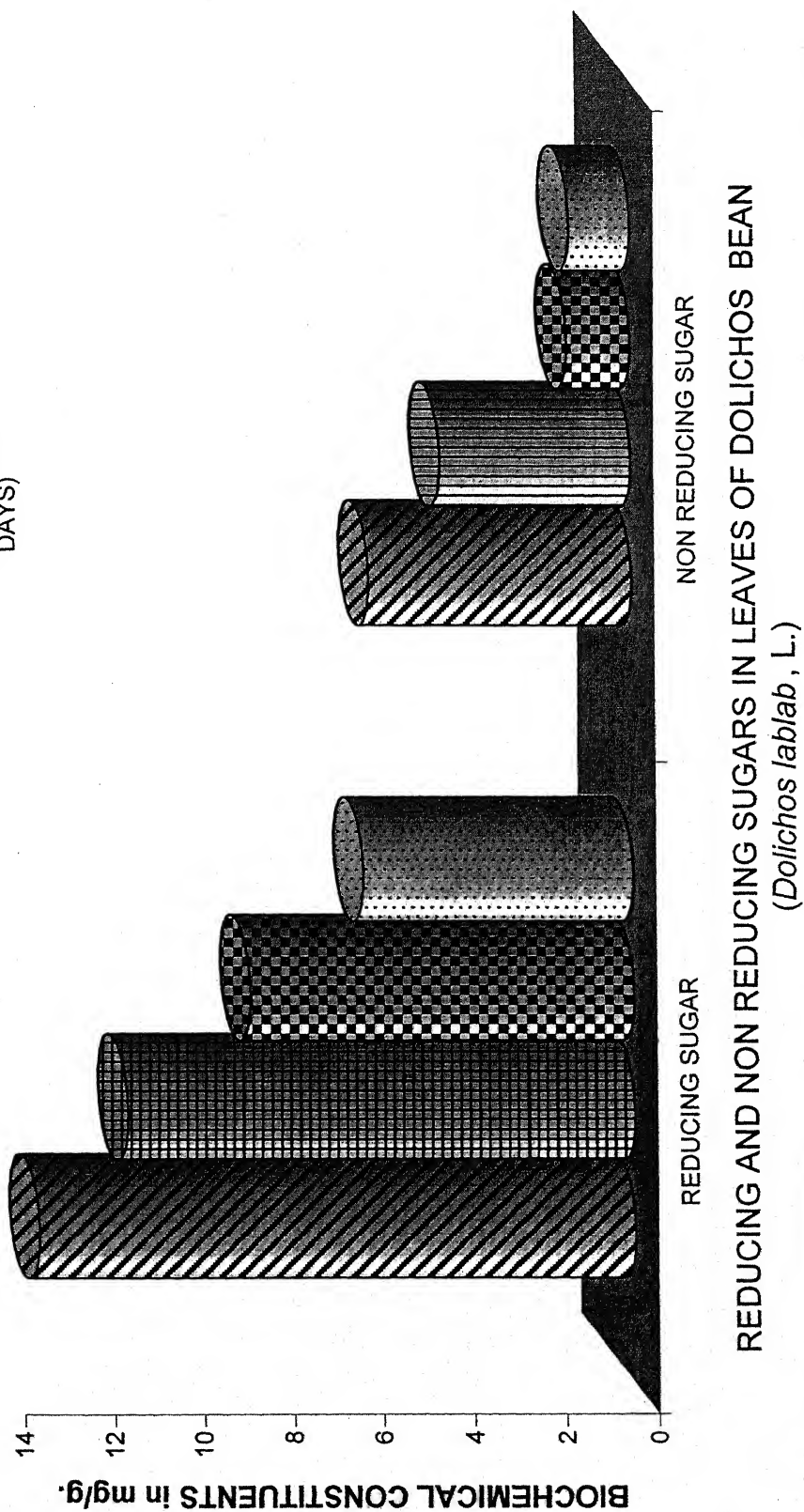
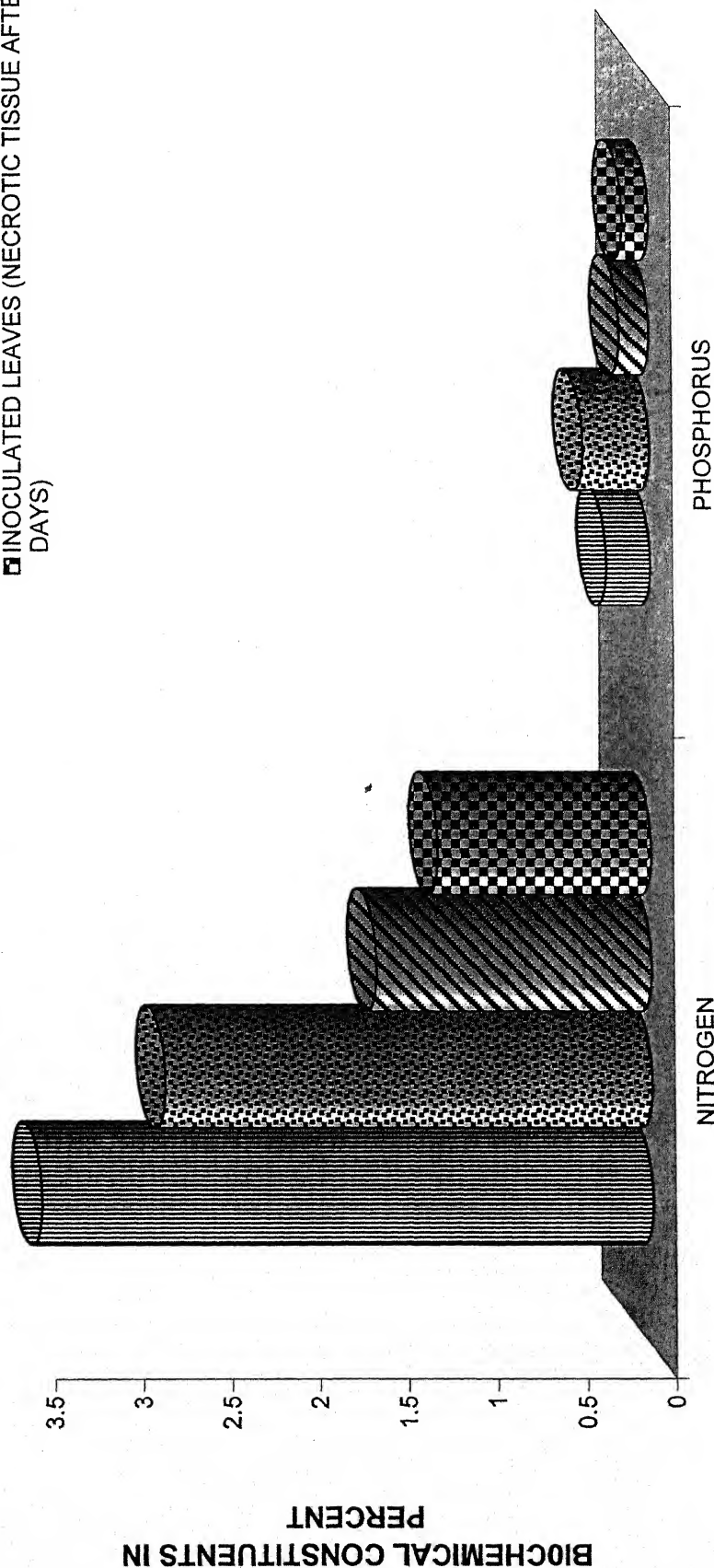


FIGURE-46

- UN-INOCULATED LEAVES
- ▨ INOCULATED LEAVES (40 DAYS)
- ▩ INOCULATED LEAVES (70 DAYS)
- ▧ INOCULATED LEAVES (NECROTIC TISSUE AFTER 70 DAYS)

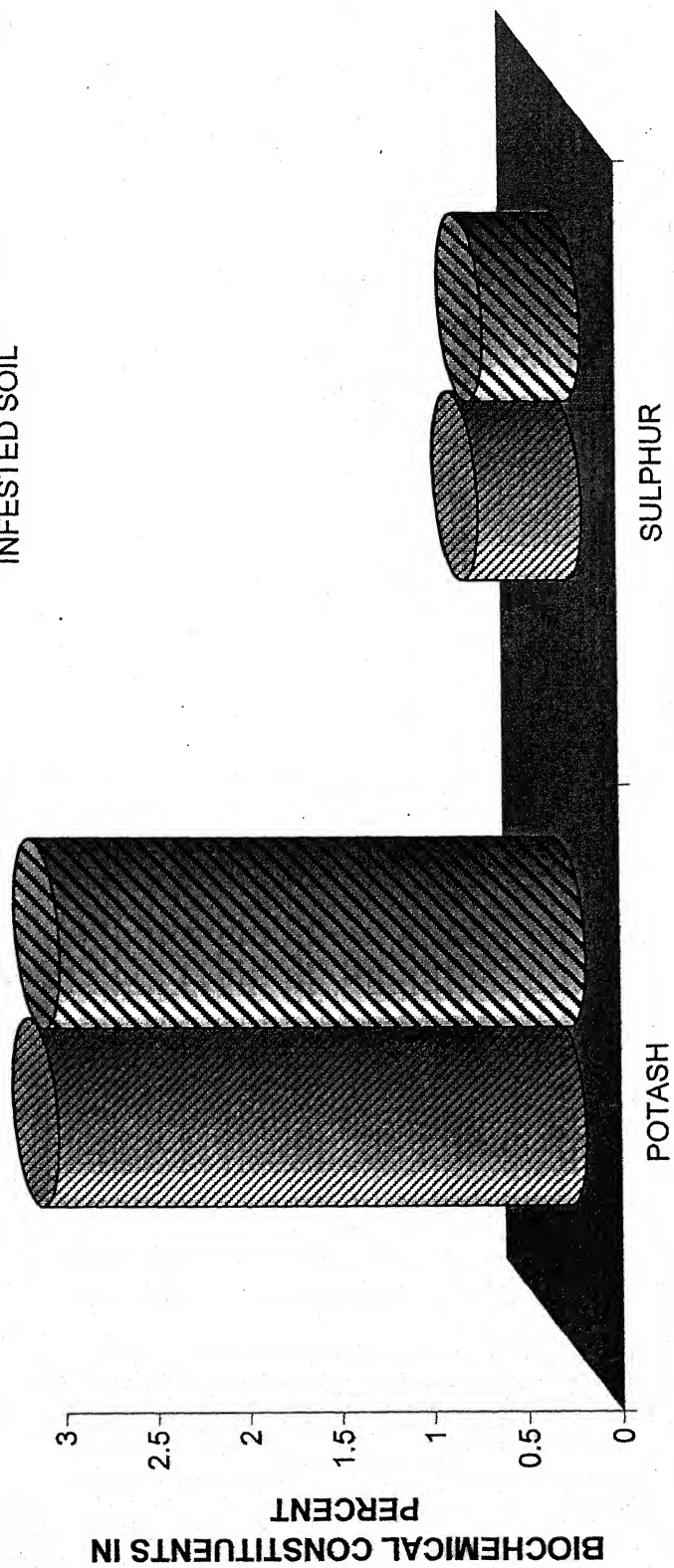


NITROGEN AND PHOSPHORUS CONTENTS IN LEAVES OF DOLICHOS BEAN
(*Dolichos lablab*, L.)

FIGURE-47

☐ STERILIZED SEEDS SOWN IN NATURALLY INFESTED SOIL

▨ STERILIZED SEEDS, SOWN IN STERILIZED INFESTED SOIL



POTASH AND SULPHUR CONTENTS IN LEAVES OF DOLICHOS BEAN
(*Dolichos lablab*, L.).

(Fries.), Keissler, isolations were made from the seeds of susceptible germplasm/culture "Kalyanpur Type - 1" of *Dolichos* bean (*Dolichos lablab*, L.) as well as from healthy seeds, mixed with spore-cum-mycelial suspension of the pathogen and the results concluded are presented in Table XXXII.

TABLE - XXXII

Role of seed in survival of *Alternaria alternata* (Fries.), Keissler, in association with *Dolichos* bean (*Dolichos lablab*, L.) seeds under laboratory conditions during the year 2001 to 2002.

Treatment	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sep.	Oct.	Nov.
Naturally infected seeds	+	+	+	+	+	+	+	+	+	+	+	+
Healthy seeds with spore suspension of <i>Alternaria alternata</i>	+	+	+	+	+	+	+	+	+	+	+	+
Sterilized healthy seeds	-	-	-	-	-	-	-	-	-	-	-	-

(+) = Denotes Survival of pathogen.

(-) = Denotes Non-survival of pathogen.

The results presented in Table XXXII revealed that seeds collected from diseased plants as well as healthy plants mixed with spore-cum-mycelial suspension of the pathogenic culture of *Alternaria alternata* (Fries.), Keissler, yielded the pathogenic culture of *Alternaria alternata*.

Thus it was concluded that both naturally infected seeds and artificially infected seeds served as a carrier of the pathogen.

(b) ROLE OF SEEDS IN THE PRIMARY INFECTION OF THE PATHOGEN IN POT CULTURE EXPERIMENT :

In order to detect out the role of seeds as a source of primary infection of the disease, observations were recorded in pots. In this experiment naturally infected diseased seeds, were sown in earthen pots containing sterilized soil. Seeds mixed with spore suspension and sterilized healthy seeds, were sown separately in pots containing sterilized soil. Plants were allowed to grow till flowering stage and protecting them from aerial infection. Plants were regularly observed for appearance of disease. The results obtained are summarised in Table XXXIII and corresponding Figures 48 and 49.

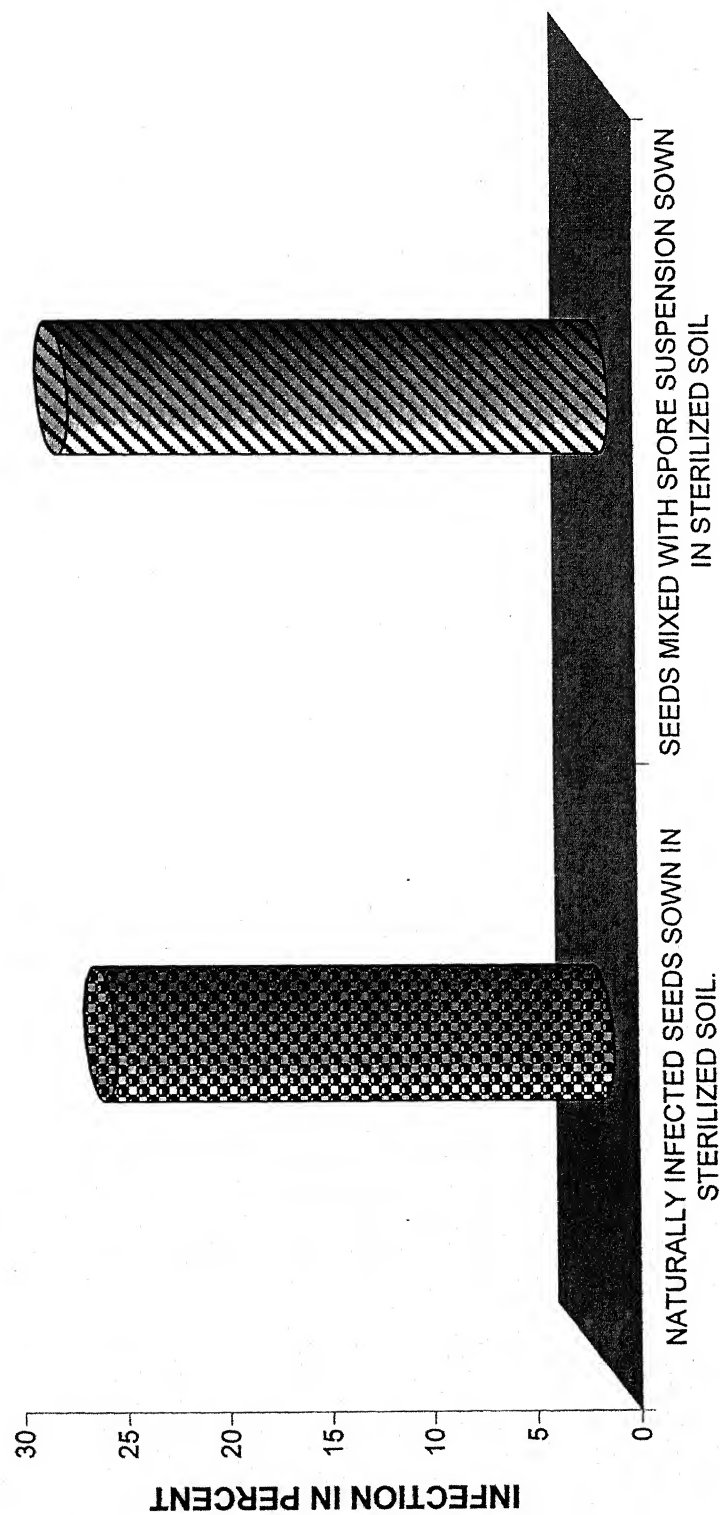
TABLE - XXXIII

Role of seeds in Primary infection of the disease leaf spot of Dolichos bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler, in Pot culture experiment.

Treatments	Number of seeds sown	Number of Plants grown	Number of Infected Plants	Infection (%)
Naturally infected seeds sown in sterilized soil	20	11	5	24.30
Seeds mixed with spore suspension and sown in sterilized soil.	20	12	6	26.50
Sterilized healthy seeds in sterilized soil (control)	20	19	—	—

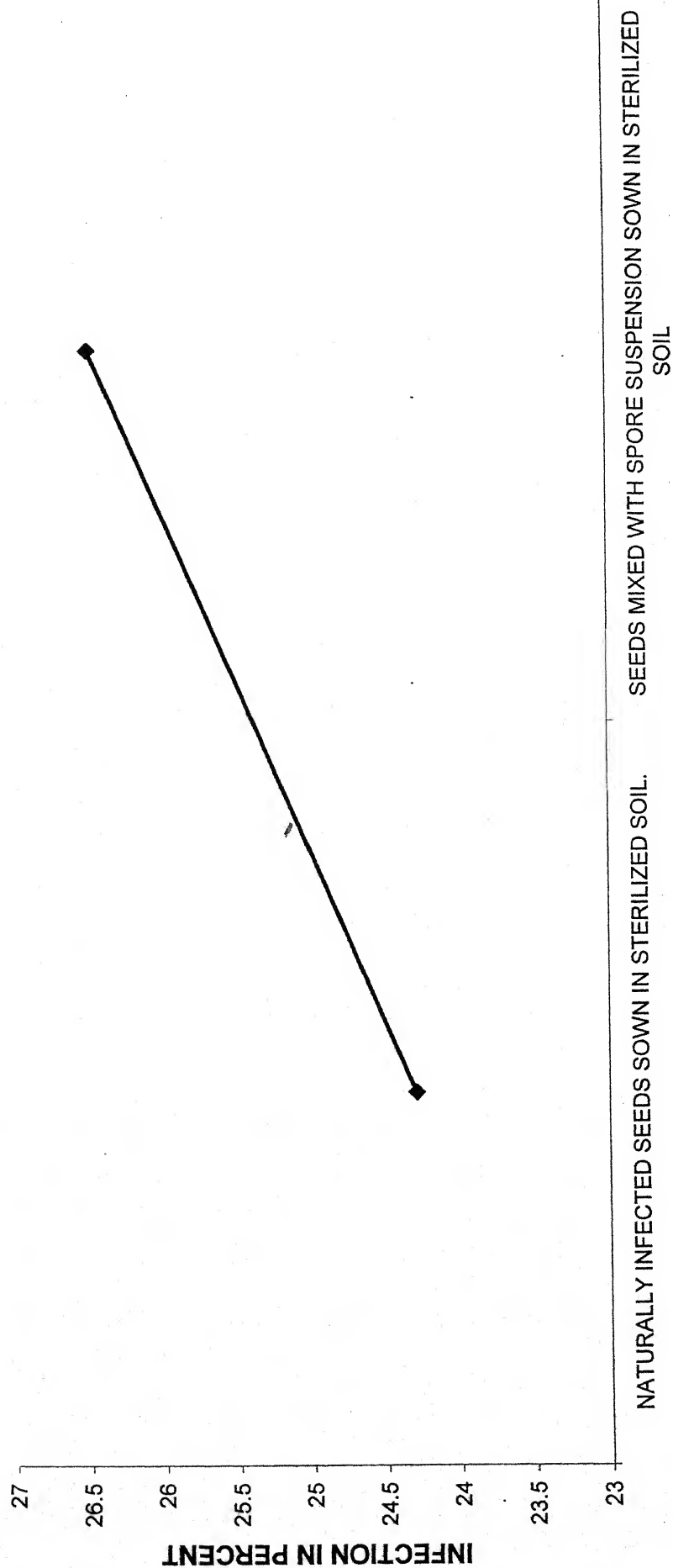
(—) = Denotes Absence of infected plants and infection.

FIGURE-48



ROLE OF SEEDS IN PRIMARY INFECTION OF THE LEAF SPOT DISEASE OF
DOLICHOS BEAN (*Dolichos lablab*, L.) IN POT CULTURE EXPERIMENT

FIGURE-49



ROLE OF SEEDS IN PRIMARY INFECTION OF THE LEAF SPOT DISEASE OF DOLICHOS
BEAN (*Dolichos lablab*, L.) IN POT CULTURE EXPERIMENT

The results presented in Table XXXIII and Figures 48 and 49 revealed, that typical symptoms of disease appeared in the plants raised from naturally infected seeds as well as artificially infested seeds. The percentage of plants infected, was recorded as higher in artificially infested seeds in comparison to that of naturally infected ones. No symptoms were observed in plants raised from healthy seeds. Thus it was proved that seeds not only played a significant role in survival of the pathogen but also in the primary infection of the disease.

2. ROLE OF SOIL :

(a) ROLE OF INFESTED SOIL IN SURVIVAL OF THE PATHOGEN :

To determine the role of infested soil as a source of primary infection the naturally infested field soil and sterilized soil mixed with fungal suspension, were stored at room temperature $25 \pm 1^{\circ}\text{C}$. The experiment was conducted as per procedure described under, "Material and Method" and data recorded are summarised in Table XXXIV.

TABLE - XXXIV

Role of infested soil in survival of pathogen *Alternaria alternata* (Fries.), Keissler, causing leaf spot of Dolichos bean (*Dolichos lablab* L.) during the years 2001 to 2002.

Treatments	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	July	Aug.	Sep.	Oct.	Nov.	Dec.
Naturally infested field soil	+	+	+	+	+	+	+	+	+	+	+	+	+
Sterilized soil infested with fungal culture	+	+	+	+	+	+	+	+	+	+	+	+	+

(+) = Denotes Pathogen isolated.

It is evident from the results presented in Table XXXIV that the pathogen, *Alternaria alternata* (Fries.), Keissler, was isolated from the naturally infested soil and artificially infested autoclaved soil from November 2001 to June 2002.

Thus it is established that the pathogen remained viable in the infested soil to infective stage from one season to another season.

(B) ROLE OF INFESTED SOIL IN PRIMARY INFECTION OF THE DISEASE :

In order to study the role of infested soil in the survival of pathogen, *Alternaria alternata* (Fries.), Keissler as primary infection of the disease, observations were taken in the pots containing naturally infested field soil, artificially infested soil and sterilized soil sown with sterilized seeds separately. Sterilized healthy seeds were sown separately in pots containing sterilized soil as control. The results obtained are summarised in Table XXXV and Figures 50 and 51.

TABLE - XXXV

Role of infested soil in Primary infection of the disease, leaf spot of *Dolichos* bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler.

Treatments	Number of seeds sown	Number of Plants grown	Number of Plants Infected	Disease Infection (%)
Sterilized seeds sown in naturally infested soil	20	15	3	20.00
Sterilized seeds, sown in sterilized infested soil	20	12	4	33.30
Sterilized seeds sown in sterilized soil	20	16	—	—

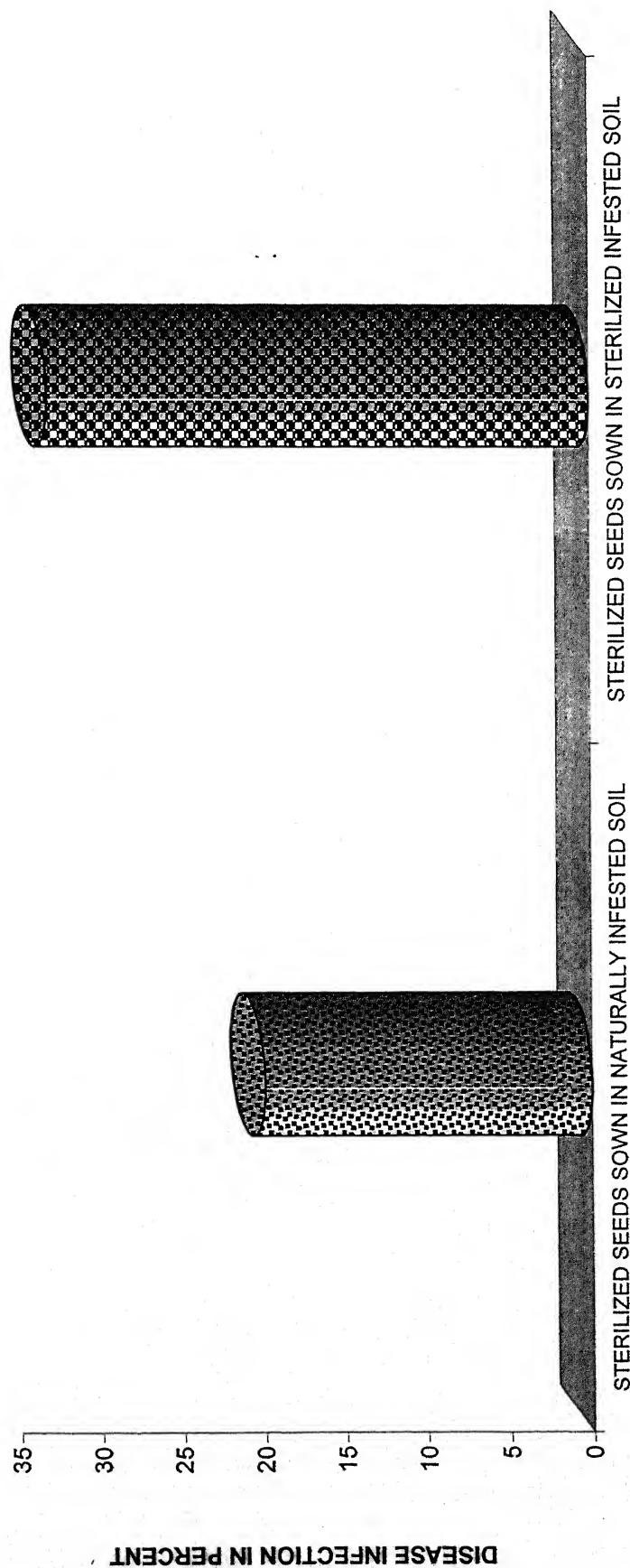
(—) = Denotes Absence of infected plants and infection.

It is obvious from the Table XXXV and Figures 50 and 51 that plants raised in the pots filled with naturally infested soil and artificially infested soil produced typical symptoms of the disease but the percentage of plant's infection was more in artificially infested soil than naturally infested field soil. No symptoms were observed, where sterilized seeds were sown in sterilized soil

FIGURE-50

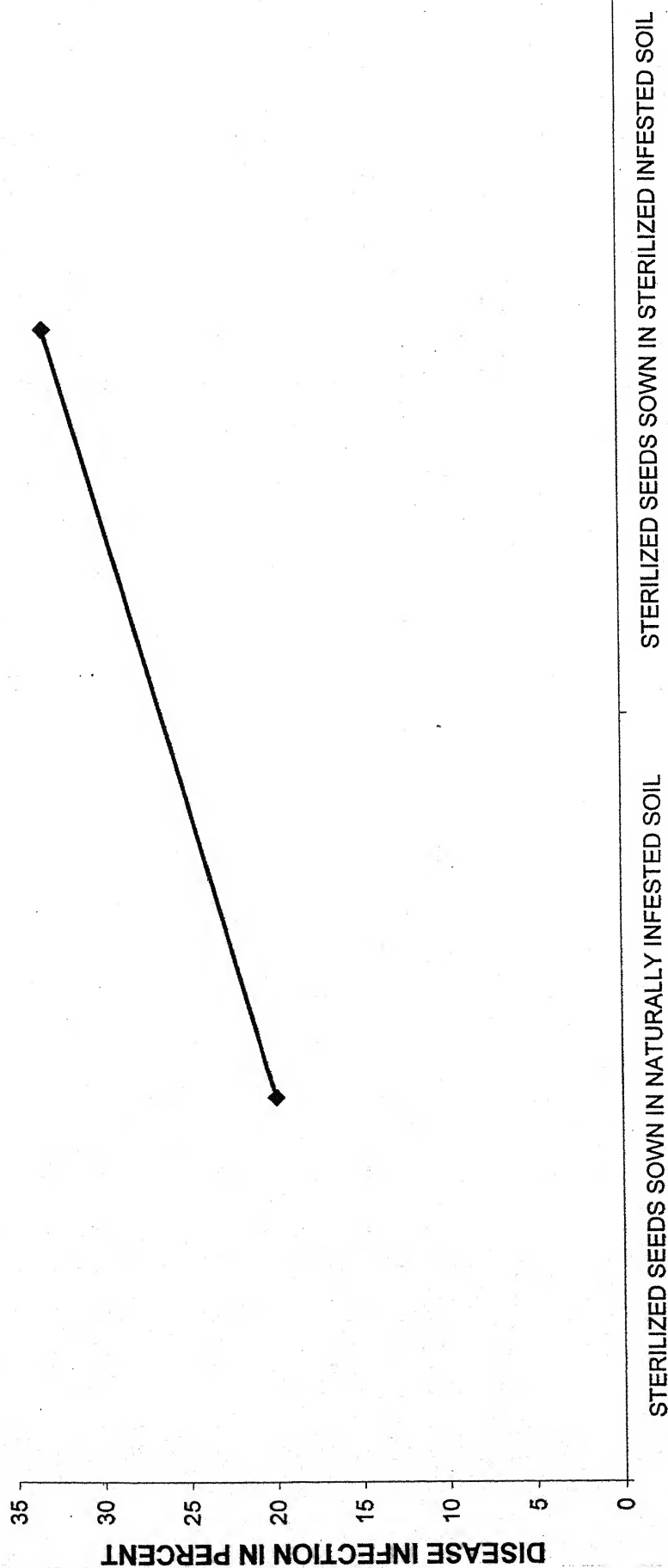
STERILIZED SEEDS SOWN IN NATURALLY INFESTED SOIL

STERILIZED SEEDS SOWN IN STERILIZED INFESTED SOIL



ROLE OF INFESTED SOIL IN PRIMARY INFECTION OF THE LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler.

FIGURE-51



ROLE OF INFESTED SOIL IN PRIMARY INFECTION OF THE LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler.

(Control). Thus results revealed that the pathogen was not only able to survive in infested soil from one season to another but it also played the role as a source of primary infection.

3. ROLE OF PLANT DEBRIS IN SURVIVAL OF THE PATHOGEN

(a) UNDER LABORATORY CONDITION :

For this study the affected leaves of highly susceptible germplasm/culture Kalyanpur Type - 1 of Dolichos bean (*Dolichos lablab*, L.), were collected from the field during the crop seasons of 2001 and 2002 and preserved in laboratory. Monthly isolations were made from the material to ascertain the survival of pathogen up to month of June 2001 (Table XXXVI). It was recorded that pathogen was able to survive on diseased leaves from November 2001 to June, 2002 in all the cases in laboratory till the next crop season and in all the cases pure culture of *Alternaria alternata* (Fries.), Keissler, was obtained. The observations recorded in Table XXXVI clearly exhibit that the pathogen remained viable in the affected leaves from one season to any other season.

TABLE-XXXVI

Survival of pathogen *Alternaria alternata* (Fries.), Keissler in diseased plant debris in laboratory and field conditions during the year 2001 and 2002.

Month/Year	Diseased plant debris in laboratory	Diseased plant debris burried in pot soil in the open
December, 2001	+	+
January, 2002	+	+
February, 2002	+	+
March, 2002	+	+
April, 2002	+	-

May, 2002	+	-
June, 2002	+	+
July, 2002	+	-
August, 2002	+	-
September, 2002	+	-
October, 2002	+	-
November, 2002	+	-

(+) = Denotes pathogen isolated.

(-) = Denotes pathogen not isolated.

(b) UNDER FIELD CONDITION :

The experiment was conducted to ascertain the role of plant debris in soil in the perpetuation of the pathogen. The diseased leaves of germplasm/culture, "Kalyanpur Type - 1" of Dolichos bean infected with *Alternaria alternata* (Fries.), Keissler, were kept in the tissue paper bags and were buried in soil at the depth of 8.0 cm. and kept in open. Subsequently regular isolations were made from the leaves at monthly intervals in order to study the role of seed viability of the pathogen as well as perpetuation of the pathogen. The pathogen was isolated till June 2002, from the buried diseased leaves but not after that, probably because of severe contamination caused by saprophytic organisms as summarised in the Table XXXVI.

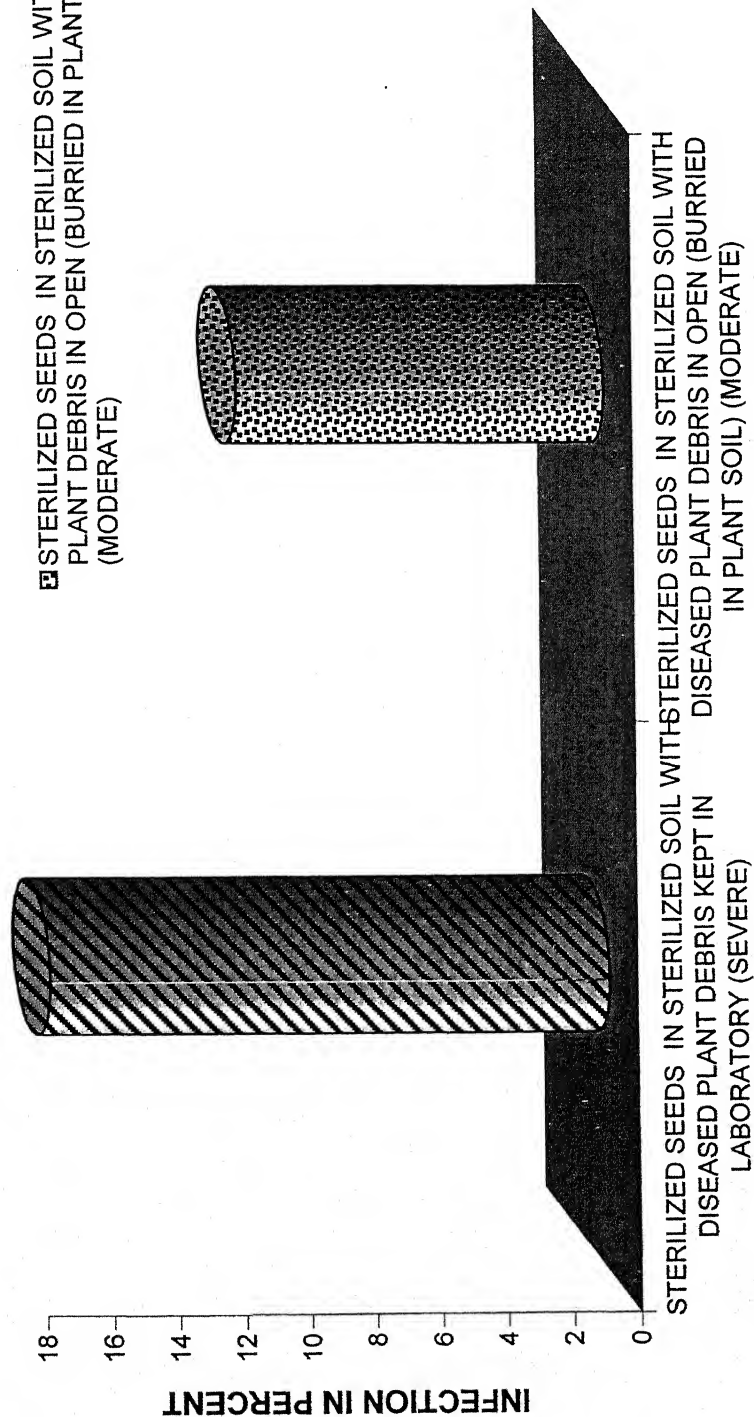
4. ROLE OF PLANT DEBRIS IN PRIMARY INFECTION OF THE PATHOGEN :

In order to study the role of diseased plant debris as a source of primary infection the five surface sterilized seeds of germplasm/culture "Kalyanpur Type-1" of Dolichos bean, were sown as suggested in "Material and Method". The observations on the development of disease symptoms, were recorded regularly and the results are summarised in Table XXXVII and Figures 52 and 53.

FIGURE-52

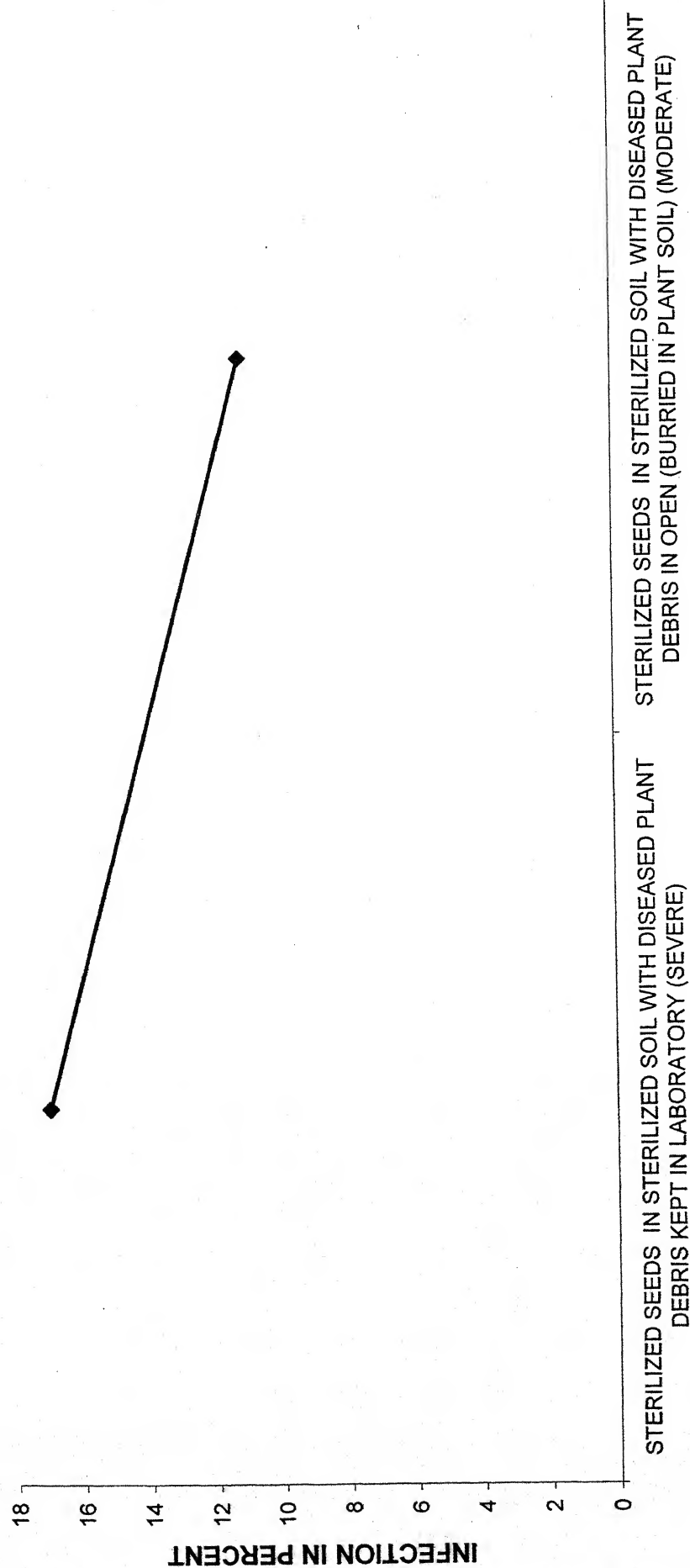
STERILIZED SEEDS IN STERILIZED SOIL WITH DISEASED PLANT DEBRIS KEPT IN LABORATORY (SEVERE)

STERILIZED SEEDS IN STERILIZED SOIL WITH DISEASED PLANT DEBRIS IN OPEN (BURRIED IN PLANT SOIL) (MODERATE)



ROLE OF DISEASED PLANT DEBRIS IN PRIMARY INFECTION OF THE
LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.)
CAUSED BY *Alternaria alternata* (Fries.), Keissler

FIGURE-53



ROLE OF DISEASED PLANT DEBRIS IN PRIMARY INFECTION OF THE LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.) CAUSED BY *Alternaria alternata* (Fries.), Keissler.

TABLE - XXXVII

Role of diseased plant debris in primary infection of the disease leaf spot of *Dolichos bean* (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler.

Treatments	Number of seeds sown	Number of plants grown	Number of plants infected	Infection (%)	Severity of Infection
Sterilized seed in sterilized soil with diseased plant debris kept in laboratory.	20	16	5	17.00	Severe
Sterilized seeds + Sterilized soil diseased plant debris in open (buried in plant soil)	20	13	3	11.25	Moderate
Sterilized seeds + sterilized soil	20	12	—	—	—

(—) = Denotes absence of infection

It is apparently clear from the results presented in Table XXXVII and Figures 52 and 53 that *Dolichos bean* plants raised in pot soil infested with plants debris stored at room temperature $25\pm 1^{\circ}\text{C}$, exhibited typical symptoms of *Alternaria* leaf spot disease. Similarly disease development also occurred in plants raised in pot soil mixed with diseased plants debris kept in open. The per cent plant infection was more in case of diseased plant debris stored in laboratory than kept in open. Thus it is established that diseased plants debris stored under field conditions played a significant role in the primary infection of the disease.

5. SECONDARY SPREAD OF DISEASE :

The experiment was conducted in order to determine the role of air borne inoculum in secondary spread of the disease. Two sets of plants were raised according to the method, described, under "Material and Method". The pots having infected plants were transferred in the vicinity of healthy plants. One set of plants was covered with muslin cloth in order to get eliminated the aerial infection, which served as control. The healthy plants were examined regularly for recording disease development and results are presented in Table XXXVIII.

TABLE - XXXVIII

Role of Air borne inoculum in secondary spread of the disease leaf spot of *Dolichos* bean (*Dolichos lablab*, L.) caused by *Alternaria alternata* (Fries.), Keissler.

Treatments	Number of Seeds sown	Number of plants grown	Number of plants infected	Disease Incidence (%)
Sterilized seeds sown in sterilized soil in pots and plants covered with muslin cloth in vicinity of diseased plants.	20	14	—	—
Sterilized seeds sown in sterilized soil in pots and plant exposed to air in vicinity of diseased plants.	20	13	5	36.30

(—) = Denotes Absence of plants infected and disease incidence.

From the results presented in Table XXXVIII, it was concluded that the pots containing the plants covered with muslin cloth did not produce the disease symptoms and escape infection, while another set which was kept uncovered and exposed to air, caused aerial infection and produced typical symptoms of leaf spot of Dolichos bean. Thus from the experiment under study, it was ascertained that the disease inoculum reached the uncovered healthy plants through the agency of air causing the infection leading to development of disease establishing the role of air borne inoculum in secondary spread of disease.

Abundant spores of the pathogen, *Alternaria alternata* (Fries.) Keissler, were also seen on the naturally infected leaves, which reached to healthy plants through agency of air.

Thus on the basis of experiment under study, it is obvious that infected seeds, soil and plant debris served as a source of primary infection and secondary spread of the disease was caused through the agency of air. Thus the *Alternaria* leaf spot of Dolichos bean incited by, *Alternaria alternata* (Fries.), Keissler, is found to be seed, soil as well as air borne in nature.

VARIETAL SCREENING FOR DISEASE RESISTANCE :

1. SCREENING OF DOLICHOS BEAN GERMPLASMS/CULTURES UNDER NATURAL CONDITIONS AGAINST *Alternaria alternata* (Fries.), Keissler :-

The use of resistant varieties is the best method to eradicate the disease. In view of this aim the present study was carried out to detect out the source of resistance against leaf spot of Dolichos bean caused by *Alternaria alternata* (Fries.), Keissler. Germplasm/cultures of Dolichos bean obtained from Vegetable Research Farm Kalyanpur, Kanpur and Chandra Shekhar Azad University of Agriculture and Technology, Kanpur, were screened under natural conditions during Kharif season of the year 2001 in order to examine their reaction to the pathogen according to the technique described under "Material and Method" and results are presented in Table XXXIX.

TABLE-XXXIX

Reaction and per cent disease intensity of Dolichos bean (*Dolichos lablab*, L.) Germplasms/Cultures to *Alternaria alternata* (Fries.), Keissler, under natural conditions during the year 2001.

S. No.	Germplasms/ Cultures	Average disease intensity in per cent	Grade	Pathogenic Reaction	Patho- genic Effect
1.	Altapati	1.71	1	+	R
2.	Arka Jai	7.25	2	+	MR
3.	Arka Vijay	—	—	—	F
4.	Culture 5508	14.35	3	+++	MS
5.	6001	18.29	3	+++	MS
6.	6009	33.81	4	+++++	HS
7.	6014	62.38	5	+++++	HS
8.	6019	58.79	5	+++++	HS
9.	6022	25.11	4	++++	S
10.	6023	24.89	4	++++	S
11.	6201	16.50	3	+++	MS
12.	6317	17.24	3	+++	MS
13.	6701	21.79	4	++++	S
14.	6801	29.27	4	++++	S
15.	6802	6.28	2	+	MR
16.	6804	13.92	3	+++	MS
17.	7001	9.13	2	+	MR
18.	7006	28.43	4	++++	S
19.	7007	27.35	4	++++	S

20.	7008 - A	15.29	3	+++	MS
	7008 - B	—	—	—	F
21.	7010	11.72	3	+++	MS
22.	7012	22.59	4	++++	S
23.	7015 - A	24.76	4	++++	S
24.	7015 - B	52.37	5	+++++	HS
25.	7016	20.54	4	++++	S
26.	7019	23.68	4	++++	S
27.	7020 - A	60.42	5	+++++	HS
28.	7020 - B	22.19	4	++++	S
29.	7022	1.45	1	+	R
30.	7005	51.49	5	+++++	HS
31.	7023	55.38	5	+++++	HS
32.	7024	16.35	3	+++	MS
33.	7101	24.92	4	++++	S
34.	7103	7.20	2	+	MR
35.	7205	23.54	4	++++	S
36.	7206	22.89	4	++++	S
37.	7207	15.49	3	+++	MS
38.	7210	17.22	3	+++	MS
39.	7301	6.39	2	+	MR
40.	7501	61.49	5	+++++	HS
41.	7601	16.42	3	+++	MS
42.	7603	23.74	4	++++	S
43.	7604	59.27	5	+++++	HS
44.	7701	14.79	3	+++	MS

45.	7702	23.57	4	++++	S
46.	7703	—	—	—	F
47.	7705	13.82	3	+++	MS
48.	7708	15.77	3	+++	MS
49.	7710	24.93	4	++++	S
50.	7711	25.13	4	++++	S
51.	8001	7.42	2	+	MR
52.	8002	6.37	2	+	MR
53.	8003	14.97	3	+++	MS
54.	8004	16.84	3	+++	MS
55.	8005	15.33	3	+++	MS
56.	8101	24.45	4	++++	S
57.	8401	27.35	4	++++	S
58.	8402	1.82	1	+	R
59.	8403	6.53	2	+	MR
60.	8405	14.67	3	+++	MS
61.	9101	—	—	—	F
62.	9102	15.49	3	+++	MS
63.	9104	26.73	4	++++	S
64.	9105	25.38	4	++++	S
65.	9108	17.29	3	+++	MS
66.	9109	16.82	3	+++	MS
67.	9110	14.52	3	+++	MS
68.	9113	15.63	3	+++	MS
69.	9114	21.37	4	++++	S
70.	9115	22.55	4	++++	S

71.	9117	12.91	3	+++	MS
72.	9118	13.78	3	+++	MS
73.	DB-1	24.29	4	++++	S
74.	DPL-1	13.72	3	+++	MS
75.	Goya	16.27	3	+++	MS
76.	HA-3	8.15	2	+	MR
77.	HD-1	6.95	2	+	MR
78.	Hatikan	23.18	4	++++	S
79.	HD-10	14.23	3	+++	MS
80.	HD-66	15.43	3	+++	MS
81.	HD - 81	8.83	2	+	MR
82.	HD - 93	59.82	5	+++++	HS
83.	HD - 104	16.35	3	+++	MS
84.	JDL - 17	1.94	1	+	R
85.	JDL - 37	—	—	—	F
86.	JDL - 79	60.93	5	+++++	HS
87.	JDL - 85	7.45	2	+	MR
88.	Kalyanpur Type - 1	58.67	5	+++++	HS
89.	Kamrang Selection - 2	55.49	5	+++++	HS
90.	Pusa Early Prolific	2.75	1	+	R
91.	Rajani	2.34	1	+	R
92.	Todi - 21	17.63	3	+++	MS
93.	Todi 125-136	—	—	—	F

(F) = Denotes Free

(+)	=	Denotes Resistant
(++)	=	Denotes Moderately Resistant
(+++)	=	Denotes Moderately Susceptible
(++++)	=	Denotes Susceptible
(+++++)	=	Denotes Highly Susceptible

It is obvious from the Table XXXIX, that six germplasms/cultures viz; Arka Vijaya, Cultures, 7703, 7708-B and 9101, JDL-37 and Todi 125-136 were found to be tolerant being Disease Free (F). The 5 germplasm/cultures viz; Altapai, Culture-7022, JDL-17, Pusa Early Prolific and Rajani, were found Resistant (R). The disease intensity varied from 1.45 - 2.75 per cent. The maximum disease intensity 2.75 per cent was found in Pusa Early Prolific followed by 2.34 per cent in Rajani. 1.94 per cent in JDL - 17, 1.71 per cent in Altapati and 1.45 per cent in culture 7022.

The 12 germplasm/cultures viz; Arka Vijai, 6802, 7001, 7103, 7301, 8001, 8002, 8403, HA-3, HD-4, HD-81 and JDL-85, were found moderately Resistant (MR). The disease intensity varied from 6.25 per cent to 9.13 per cent. The maximum disease intensity 9.13 per cent was found in culture-7001, followed by 8.83 per cent in HD-81; 8.15 per cent in HA-3; 7.45 per cent in JDL-85; 7.42 per cent in culture-8001, 7.25 per cent in Arka Jai; 7.20 per cent in culture 7103; 6.95 per cent in HD-1; 6.53 per cent in culture 8002; 6.39 per cent in culture 7301 and 6.28 per cent in culture - 6802.

The 31 germplasms/ cultures viz; 5508, 6001, 6201, 6317, 6804, 7008-A, 7010, 7024, 7027, 7210, 7601, 7701, 7705, 7708, 8003, 8004, 8005, 8405, 9102, 9108, 9109, 9110, 9113, 9117, 9118, DPL-1, Goya, HD-10, HD-66, HD-104 and Todi - 21, were found Moderately susceptible (MS). The disease intensity was found varied from 11.72 per cent to 18.29 per cent. The maximum disease intensity 18.29 per cent was found in culture - 6001, followed by 17.63 per cent in Todi - 21; 17.29 per cent in culture - 9108; 17.24 per cent in culture 6317; 17.22 per cent in culture - 7210; 16.84 per cent in culture-8004; 16.82 per cent in culture - 9108; 16.50 per cent in culture-6201; 16.42 per cent in culture-7601; 16.35 percent in HD-104 and culture 7024; 16.27 per cent in Goya; 15.77 per

cent in culture-7708; 15.63 per cent in culture-9113; 15.49 per cent in culture 7207; and 15.43 per cent in HD-66 and 9102; 14.97 percent in culture-8003; 15.33 per cent in culture-8005; 15.29 per cent in culture-7008 A; 14.79 per cent in culture-7701; 14.67 per cent in culture-8405; 14.52 per cent in culture-9110; 14.35 percent in culture-5508; 14.23 per cent in HD-10; 13.92 per cent in culture-6804; 13.82 per cent in culture-7705; 13.78 per cent in culture-9118; 13.72 per cent in culture DPL-1; 12.91 per cent in culture 9117 and 11.72 per cent in culture 7010.

The 26 permplasms/cultures viz; 6022, 6023, 6701, 6801, 7006, 7007, 7012, 7015, 7016, 7019, 7020, 7101, 7205, 7206, 7603, 7702, 7710, 7711, 8101, 8401, 9104, 9105, 9114, 9116, DB-1 and Hatikan, were found susceptible (S.). The disease intensity was found varied from 20.54 per cent to 29.27 per cent. The maximum disease intensity 29.27 per cent was found in culture 6801, followed by 28.43 per cent in culture-7006; 27.35 per cent in culture-8401 and 7007; 26.73 per cent in culture-9104; 25.38 per cent in culture-9105; 25.18 per cent in culture-2711; 25.11 per cent in culture-6022; 24.92 per cent in culture-7710; 24.89 per cent in culture 6023; 24.76 per cent in culture-7015; 24.45 per cent in culture-8101; 24.19 per cent in DB-1; 23.74 per cent in culture-7603; 23.68 per cent in culture 7019; 23.57 per cent in culture 7702; 23.54 per cent in culture-7205; 23.18 per cent in Hatikan; 22.59 per cent in culture 7012; 22.55 per cent in culture-9116; 22.19 per cent in culture 7020-B; 21.79 per cent in culture-6701; 21.37 per cent in culture-9114 and 20.54 per cent in culture-7016.

Thirteen germplasms/cultures viz, 6009, 6014, 6019, 7005, 7015-B, 7020-A, 7023, 7501, 7604, HD-93, JDL-79, Kalyanpur type - 1 and Kamgranj Selection - 2 were found Highly susceptible (H-5). The disease intensity varied from 51.49-62.38 per cent. The maximum disease intensity 62.38 per cent was found in culture 6014, followed by 61.49 per cent in culture 7501; 60.93 per cent in JDL-79; 60.42 per cent in culture-7020; 59.82 per cent in HD-93; 59.27 per cent in culture-7604; 58.67 per cent in Kalyanpur type - 1; 55.49 per cent in

Kamgranj selection - 2, 55.38 per cent in culture-7023; 53.81 per cent in culture-6009; 52.37 per cent in culture-7015-B and 51.49 per cent in culture-7005.

2. REACTION OF DOLICHOS BEAN GERMPLASMS/CULTURES UNDER ARTIFICIAL EPIPHYTOTIC CONDITIONS OF INOCULATION AGAINST *Alternaria alternata* (Fries), Keissler.

Twenty two germplasm/cultures, which were found diseased Free (F), Resistant (R) and Moderately resistant (MR), were further screened under artificial conditions of inoculation for their reaction to pathogen during season of the year 2001 by the technique described under "Material and Method", the observations on the disease intensity, were recorded and the results obtained are summarised in Tables XXXX and XXXXI.

(A) BY INOCULATION OF SEEDS :-

The seeds of different germplasms/cultures, were screened by inoculating with the pathogen, *Alternaria alternata* (Fries.), Keissler and the disease symptoms were studied as well as the disease intensity was recorded and the results obtained are summarised in Table XXXX.

TABLE - XXXX

Reaction and Per cent disease intensity of Dolichos bean (*Dolichos lablab*, L.) Germplasms/Cultures to *Alternaria alternata* (Fries.), Keissler, under artificial epiphytotic conditions by inoculation of seeds, during the year 2001.

S. No.	Germplasms/ Cultures	Average disease intensity in per cent	Grade	Pathogenic Reaction	Pathogenic Effect
1.	Altapati	10.36	3	+++	MS
2.	Arka Jai	13.20	3	+++	MS

3.	Arka Vijaya	5.80	2	+	MR
4.	Culture-6802	11.29	3	+++	MS
5.	7001	8.43	2	+	MR
6.	7022	8.25	2	+	MR
7.	7103	24.57	4	++++	S
8.	7301	7.63	2	+	MR
9.	7703	28.78	4	++++	S
10.	7708-B	18.59	3	+++	MS
11.	8001	28.49	4	++++	S
12.	8002	5.94	2	+	MR
13.	8403	23.82	4	++++	S
14.	9101	9.45	2	+	MR
15.	DDL-37	15.28	3	+++	MS
16.	HA-3	7.20	2	+	MR
17.	HD-4	18.19	3	+++	MS
18.	HD-81	5.87	2	+	MR
19.	JDL-17	14.38	3	+++	MS
20.	JDL-37	25.11	4	++++	S
21.	JDL-85	27.64	4	++++	S
22.	Rajani	8.35	3	+++	MR
23.	Todi 125-126	7.85	2	+	MR

(++) = Denotes Moderately Resistant

(+++)= Denotes Moderately Susceptible

(++++)= Denotes Susceptible

The results concluded in Table XXXX exhibited that none of Dolichos bean germplasm/culture was found to be immune and resistant. Out of 22 germplasms/cultures in seed inoculation test ten germplasms/cultures viz; Arka Vijai, Cultures viz; 6802, 7022, 7301, 8002 and 9001, HA-3, HD-81, Rajani and Todi 125-126 were found Moderately Resistant (MR.) The disease intensity varied from 5.80 per cent to 9.45 per cent. The maximum disease intensity 9.45 per cent was recorded in culture 9101, followed by 8.43 per cent in Rajni, 8.25 per cent in culture-7022, 7.85 per cent in Todi 125-126, 7.63 per cent in culture-7301, 7.20 per cent in HA-3, 5.94 per cent in culture-8002; 5.87 per cent in HD-81 and 5.80 per cent in Arka Vijaya.

The seven germplasms/cultures viz; Altapati, Arka Jai, Cultures-6802, 7708-B, DDL-37, HD-4 and JDL-17 were found Moderately susceptible (MS). The disease intensity varied from 10.36 to 18.59 per cent. The maximum disease intensity 18.59 per cent was observed in culture 7708-B, followed by 18.19 per cent in HD-4, 15.28 per cent in DDL-37, 14.38 per cent in JDL-17, 13.20 per cent in Arka Jai. 11.29 per cent in culture 6802 and 10.36 per cent in Altapati.

The remaining six germplasms/cultures viz; 7103, 8001, 8403, 7703, JDL-85 and DL-37, were found susceptible (S). The disease intensity varied from 23.82 per cent to 28.80 per cent. The maximum disease intensity 28.8 per cent was observed in culture-7703, followed by 28.49 per cent in culture-8001, 27.64 per cent in JDL-85, 25.11 per cent in DL-37, 24.57 per cent in culture-7103 and 23.82 per cent in culture-8403.

(B) BY INOCULATION OF POTTED PLANTS :-

The plant inoculation method was employed to study the disease symptoms caused by *Alternaria alternata* (Fries.), Keissler, as well as to control the disease. In view of this the seeds of different germplasms/cultures, were further screened by the method described under "Material and Method". The observations regarding the disease intensity were recorded and the results obtained are summarised in Table XXXXI.

TABLE - XXXXI

Reaction and Per cent disease intensity of Dolichos bean (*Dolichos lablab*, L.) Germplasms/Cultures to *Alternaria alternata* (Fries.), Keissler, under artificial epiphytotic conditions by inoculation of potted plants during the year, 2001.

S. No.	Germplasms/Cultures	Average disease intensity in per cent	Grade	Pathogenic Reaction	Pathogenic Effect
1.	Altapati	12.47	3	+++	MS
2.	Arka Jai	7.19	2	+	MR
3.	Arka Vijaya	12.84	3	+++	MS
4.	Culture-6802	15.37	3	+++	MS
5.	7001	9.16	2	+	MR
6.	7022	6.73	2	+	MR
7.	7103	21.54	4	++++	S
8.	7301	12.78	3	+++	MS
9.	7703	29.67	4	++++	S
10.	7708-B	27.56	4	++++	S
11.	8001	18.29	3	+++	MS
12.	8002	15.64	3	+++	MS
13.	8403	23.86	4	++++	S
14.	9101	8.93	2	+	MR
15.	DDL-37	8.34	2	+	MR
16.	HA-3	14.39	3	+++	MS
17.	HD-4	6.28	2	+	MR
18.	HD-81	24.78	4	++++	S
19.	JDL-17	11.92	3	+++	MS

20.	JDL-37	17.89	3	+++	MS
21.	JDL-85	7.69	2	+	MR
22.	Rajani	9.15	2	+	MR
23.	Todi 125-126	8.47	2	+	—

(++) = Denotes Moderately Resistant.

(+++)= Denotes Moderately Susceptible.

(++++)= Denotes Susceptible.

It is obvious from the Table XXXXI that nine germplasms/cultures viz; Arka Jai, cultures-7001, 7022, 9101, DDL-37, HD-4, JDL-85, Rajani and Todi 135-136 were found Moderately Resistant (MR.). The disease intensity varied from 6.28 per cent to 9.16 per cent. The maximum disease intensity 9.16 per cent was observed in culture-7001, followed by 9.15 per cent in Rajani; 8.93 per cent in culture-9101; 8.47 per cent in Todi-125-126, 8.34 per cent in DDL-37, 7.69 per cent in JDL-85; 7.19 per cent in Arka Jai and 6.73 per cent in culture-7022, while minimum 6.28 per cent in HD-4.

The nine germplasms/cultures viz; Altapati, Arka Vijaya, cultures 6802, 7301, 8001, 8002, HA-3, JDL-17 and JDL-37 were found Moderately Susceptible (MS). The disease intensity varied from 11.92 per cent to 18.29 per cent. The maximum disease intensity 18.29 per cent, was observed in culture-8001; followed by 17.89 per cent in JDL-37; 15.64 per cent in culture-8002; 15.37 per cent in culture-6802; 14.39 per cent in HA-3; 12.84 per cent in Arka Vijaya; 12.78 per cent in culture-7301 and 12.47 per cent in Altapati, while minimum 11.92 per cent in JDL-17.

The remaining five germplasms/cultures viz; cultures 7103, 7793, 7708-B, 8403 and HD-81, were found susceptible (S) under artificial conditions of inoculation. The disease intensity varied from 21.54 per cent to 29.67 per cent. The maximum disease intensity 29.67 per cent was observed in culture-7703; followed by 27.56 per cent in 7708-B; 24.78 per cent in HD-81 and 23.86 per cent

in culture 8403, while minimum 21.54 per cent in culture-7103.

The germplasms/cultures, which showed resistance under natural conditions became susceptible under artificial conditions of inoculations.

HOST RANGE OF THE PATHOGEN :

Host range of the pathogen, plays, an important role in the recurrence of the disease, leaf spot of Dolichos bean. The experiment was conducted to know the ability of pathogen. *Alternaria alternata* (Fries.), Keissler causing leaf spot of Dolichos bean to infect the different plant species including crops and weeds. For this study 70 host plants belonging to 19 different families, viz., Apocynaceae, Aracaceae, Asteraceae, Brassicaceae, Chenopodiaceae, Cucurbitaceae, Euphorbiaceae, Fabaceae, Lamiaceae, Liliaceae, Linaceae, Malvaceae, Myrtaceae, Papaveraceae, Pedaliaceae, Poaceae, Rosaceae, Solanaceae and Umbelliferae (Apiaceae), were artificially inoculated with the spore-cum-mycelial suspension of the pathogen, *Alternaria alternata* (Fries.) Keissler, as described under "Material and Method". The inoculated plants were kept in moist chamber for 48 hours and during the course they were kept moist by spraying sterilized water to provide sufficient humidity and symptoms were recorded after 15 days of inoculation, summarised in Table XXXXII and Plates No. V, VI, VII, VIII and IX.

TABLE - XXXXII

Reaction of different plant species to *Alternaria alternata* (Fries.), Keissler, under artificial condition of inoculation.

S.No.	Botanical Name	Host Plants		Pathological Reaction
		Common Name	Family	
1.	<i>Chenopodium album</i>	Bathua	Chenopodiaceae	+
2.	<i>Beta vulgaris</i>	Chukandar	" "	+
3.	<i>Spinacea oleracea</i>	Spinach	" "	-

4.	<i>Carthamus tinctorius</i>	Safflower	Asteraceae	+
5.	<i>Tagetes erecta</i>	Marigold	" "	+
6.	<i>Chrysanthemum indicum</i>			+
7.	<i>Dahlia sp.</i>			+
8.	<i>Launea asplenifolia</i>	Launea	" "	-
9.	<i>Helianthus annuus</i>	Sunflower	" "	+
10.	<i>Lactuca sativa</i>	Lettuce	" "	-
11.	<i>Brassica campestris</i>	Mustard (Sarson)	Brassicaceae	+
12.	<i>B. campestris</i> <i>var. dichotoma</i>	Black Mustard	" "	+
13.	<i>B. juncea</i>	Rye	" "	+
14.	<i>B. oleracea var. botrytis</i>	Cauliflower	" "	+
15.	<i>B. oleracea var. capitata</i>	Cabbage	" "	+
16.	<i>B. oleracea var.</i> <i>gongylodes</i>	Knolkhol	" "	+
17.	<i>Raphanus sativus</i>	Radish	" "	+
18.	<i>Cucurbita maxima</i>	Pumpkin	Cucurbitaceae	+
19.	<i>Lagenaria vulgaris</i>	Bottle Gourd	" "	+
20.	<i>Cucumis melo</i>	Kharbuza	" "	-
21.	<i>C. sativus</i>	Khira	" "	-
22.	<i>Citrullus vulgaris</i>	Tinda	" "	-
23.	<i>Luffa cylindrica</i>	Ghia Torai	" "	+

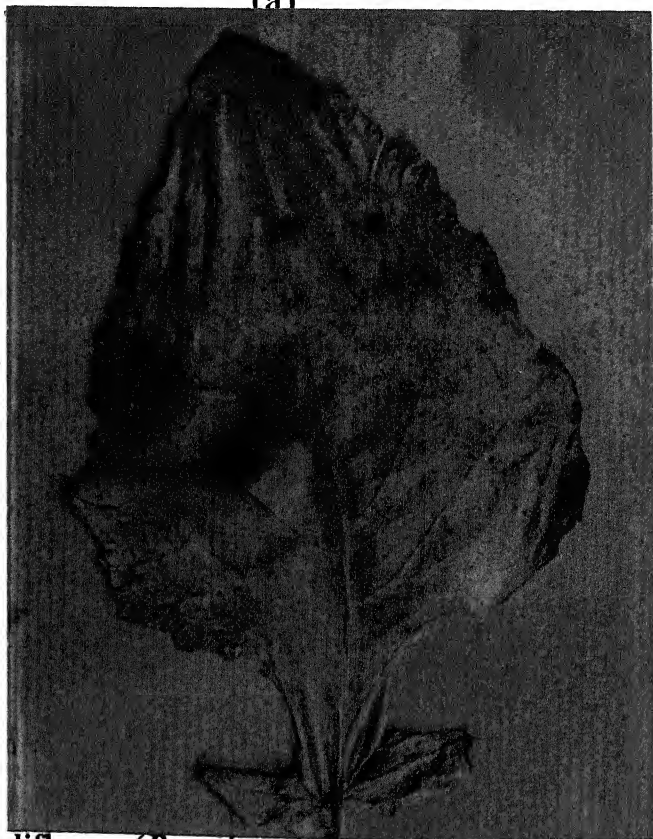
24.	<i>Euphorbia thymifolia</i>	Small Duddhi	Euphorbiaceae	—
25.	<i>Croton tiglium</i>	Jamalgota	" "	—
26.	<i>Ricinus communis</i>	Castor	" "	+
27.	<i>Avena sativa</i>	Oat	Poaceae	+
28.	<i>Cynodon dactylon</i>	Doobgrass	" "	+
29.	<i>Hordeum vulgare</i>	Barley	" "	+
30.	<i>Triticum aestivum</i>	Wheat	" "	+
31.	<i>Oryza sativa</i>	Paddy	" "	—
32.	<i>Pennisetum typhoides</i>	Bajra	" "	+
33.	<i>Sorghum vulgare</i>	Jowar	" "	+
34.	<i>Saccharum officinarum</i>	Sugarcane	" "	+
35.	<i>Zea mays</i>	Maize	" "	—
36.	<i>Cymbopogon flexuosus</i>	Lemongrass	" "	—
37.	<i>Pisum sativum</i>	Pea	Fabaceae	+
38.	<i>Cicer arietinum</i>	Gram	" "	—
39.	<i>Phaseolus radiatus</i>	Urd	" "	—
40.	<i>P. mungo</i>	Mung	" "	—
41.	<i>Cajanus cajan</i>	Arhar	" "	+
42.	<i>Arachis hypogea</i>	Groundnut	" "	+
43.	<i>Glycine max</i>	Soyabean	" "	+
44.	<i>Lens esculenta</i>	Masoor	" "	—
45.	<i>Crotolaria juncea</i>	Sunn	" "	+

46.	<i>Trifolium alexandrinum</i>	Barseem	" "	—
47.	<i>Abelmoschus esculentus</i>	Bhindi	Malvaceae	+
48.	<i>Althea rosea</i>	Hollyhock	" "	+
49.	<i>Abutilon indicum</i>	Kanghi	" "	+
50.	<i>Sida acuta</i>	Sida	" "	+
51.	<i>Hibiscus rosa-sinensis</i>	Gurhal	" "	+
52.	<i>Latana aculeata</i>	Langana	Myrtaceae	—
53.	<i>Sesamum indicum</i>	Til	Pedaliaceae	+
54.	<i>Rosa sinensis</i>	Rose	Rosaceae	—
55.	<i>Solanum nigrum</i>	Makoi	Solanaceae	+
56.	<i>Solanum tuberosum</i>	Potato	" "	+
57.	<i>Solanum melongena</i>	Brinjal	" "	+
58.	<i>Capsicum annum</i>	Lal Mirch	" "	+
59.	<i>Datura alba</i>	Dhatura	" "	+
60.	<i>Lycopersicum esculentum</i>	Tomato	" "	+
61.	<i>Solanum xanthocarpum</i>	Bhatkatayya	" "	+
62.	<i>Linum usitatissimum</i>	Linseed	Linaceae	+
63.	<i>Allium cepa</i>	Onion	Liliaceae	+
64.	<i>Coriandrum sativum</i>	Coriander	Umbelliferae (Apiaceae)	+
65.	<i>Daucus carota</i>	Gajar	" "	—
66.	<i>Argemone mexicana</i>	Pilikateri	Papaveraceae	+
67.	<i>Carissa carandus</i>	Karonda	Apocynaceae	+

PLATE -V

Host Range of Pathogen *Alternaria alternata* (Fries.), Keissler

(a)



Cauliflower (*Brassica oleracea* var. *botrytis*)

(b)



Sarson (*Brassica campestris*)

PLATE -VI

Host Range of Pathogen *Alternaria alternata* (Fries.), Keissler

(a)



Radish (*Raphanus sativus*)

(b)

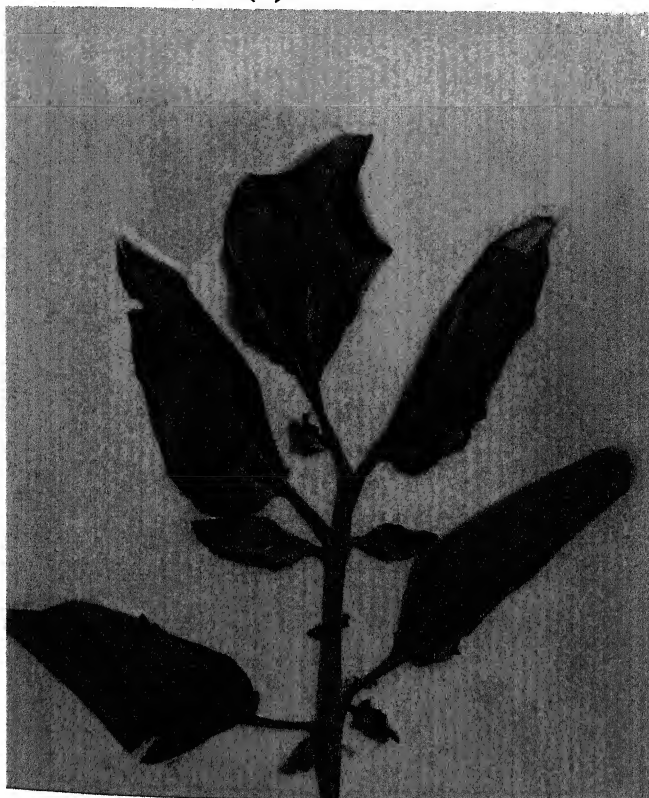


Chukandar (*Beta vulgaris*)

PLATE -VII

Host Range of Pathogen *Alternaria alternata* (Fries.), Keissler

(a)



Potato (*Solanum tuberosum*)

(b)

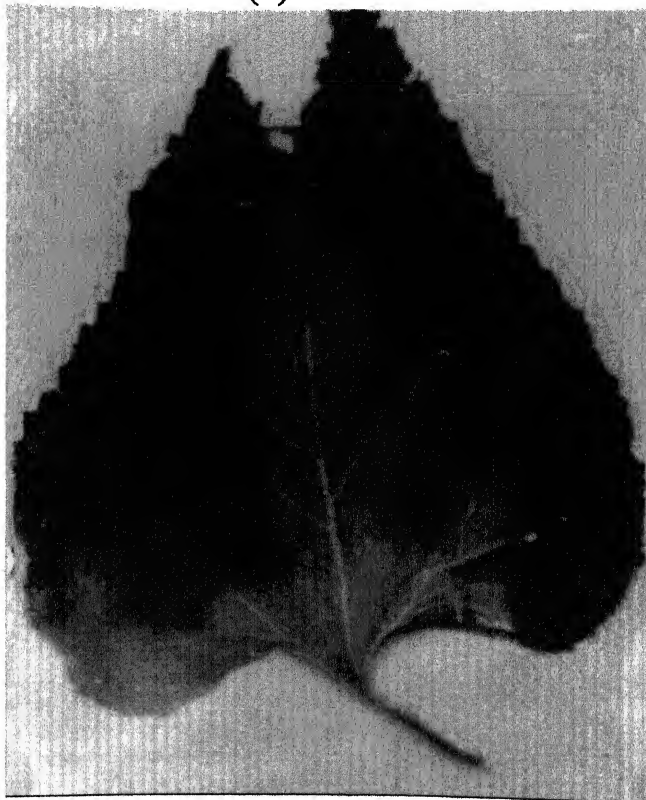


Castor (*Ricinus communis*)

PLATE -VIII

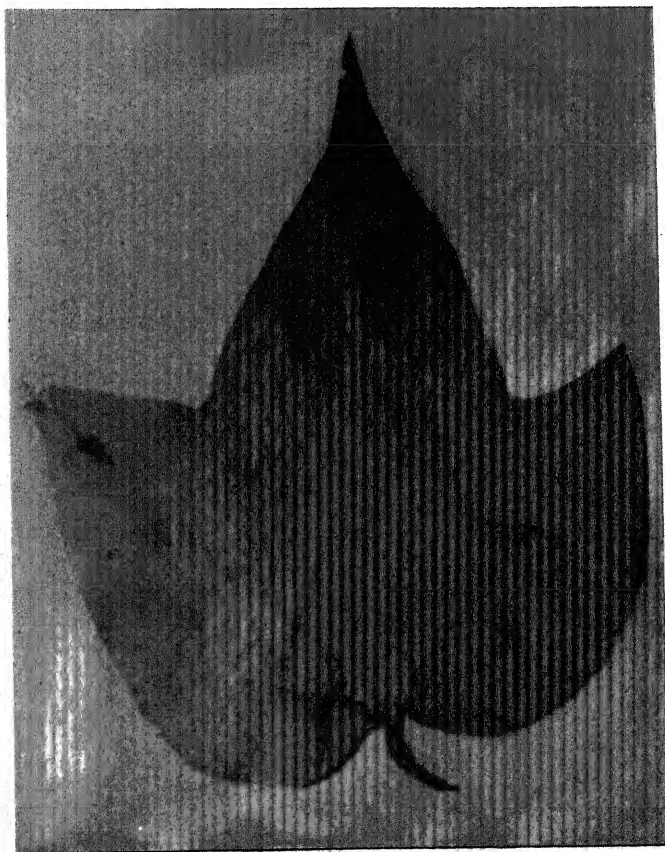
Host Range of Pathogen *Alternaria alternata* (Fries.), Keissler

(a)



Sunflower (*Helianthus annuus*)

(b)

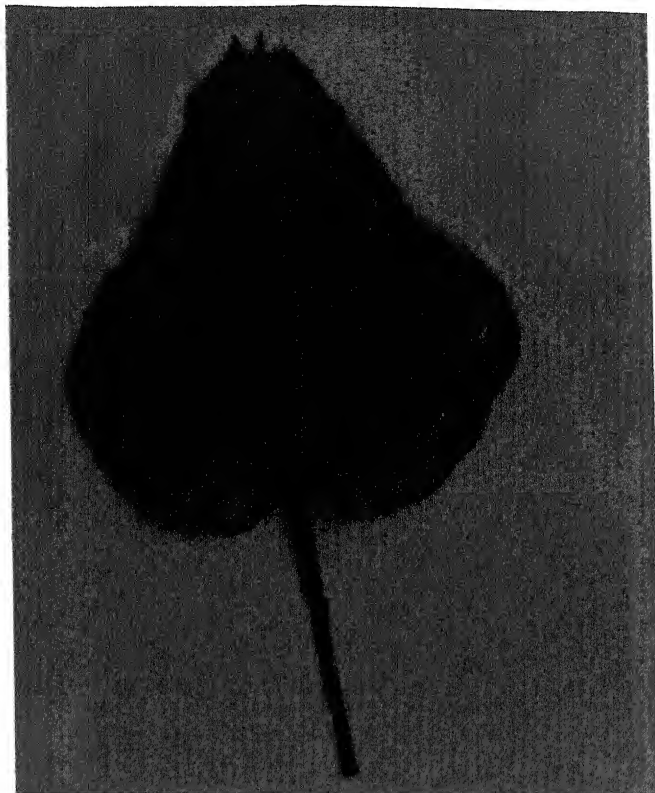


Cotton (*Gossypium herbaceum*)

PLATE -IX

Host Range of Pathogen *Alternaria alternata* (Fries.), Keissler

(a)



Brinjal (*Solanum melongena*)

(b)



Capsicum (*Capsicum annuum*)

68.	<i>Thevetia nerifolia</i>	—	" "	—
69.	<i>Ocimum sanctum</i>	Tulsi	Lamiaceae	+
70.	<i>Colocasia antiquorum</i>	Colocasia	Aracaceae	+

(+) = Denotes presence of pathological reaction.

(-) = Denotes absence of pathogenic reaction.

It is obvious from the Table XXXXII and Plates No. IV, V and VI that *Alternaria alternata* (Fries.), Keissler, was able to infect *Abelmoschus esculentus*, *Abutilon indicum*, *Althea rosea*, *Allium cepa*, *Argemone mexicana*, *Avena sativa*, *Arachis hypogea*, *Brassica campestris*, *B. campestris* var. *dichotoma*, *B. juncea*, *B. oleracea*, Var. *botrytis*, *B. oleracea* Var. *Capitata*, *B. oleracea* Var. *gongylodes*, *Carrica carandus*, *Cajanus cajan*, *Capsicum annuum*, *Carthamus tinctorius*, *Chenopodium album*, *Chrysanthemum indicum*, *Colocasia antiquorum*, *Coriandrum sativum*, *Crotolaria juncea*, *Cucurbita maxima*, *Cynodon dactylon*, *Dahlia* sp., *Datura alba*, *Glycine max*, *Helianthus annuus*, *Hibiscus rosa-sinensis*, *Hordeum vulgare*, *Lagenaria vulgaris*, *Linum usitatissimum*, *Lycopersicum esculentum*, *Luffa cylindrica*, *Ocimum sanctum*, *Pennisetum typhoides*, *Pisum sativum*, *Raphanus sativus*, *Ricinus communis*, *Saccharum officinarum*, *Sesamum indicum*, *Solanum nigrum*, *S. melongena*, *S. tuberosum*, *S. xanthocarpum*, *Sorghum vulgare*, *Tagetes erecta* and *Triticum aestivum* belonging to 17 different families viz., Apocynaceae, Aracaceae, Asteraceae, Brassicaceae, Chenopodiaceae, Cucurbitaceae, Euphorbiaceae, Fabaceae, Lamiaceae, Liliaceae, Linaceae, Malvaceae, Pedaliaceae, Papaveraceae, Poaceae, Solanaceae and Umbelliferae.

The pathogen, *Alternaria alternata* (Fries.), Keissler, was reisolated from the aforesaid inoculated hosts. It is therefore, evident that pathogen is not host specific, but has a wide host range.

SCREENING OF FUNGITOXICANTS AGAINST THE PATHOGEN IN VITRO :

Twenty five fungicides belonging to different groups (Benzene, Copper compound, Dithiocarbamate, Heterocyclic nitrogen compound, Organomercurials, Quinone, Sulphur compound, Systemic and an antibiotic viz., Agrosan G.N., Bavistin, Benlate, Blitox-50, Brassicol, Calixin, Captan, Ceresan (Dry), Dichlone, Dithane M-45, Dithane Z-78, Duter, Emisan-6, Ferbam, Foltaf 80-W, Hexaferb, Karathane, Kavach, Pancotine, Ridomil, Spergon, Suflex , Thiram, Vitavax and Ziram and antibiotic (Aureofungin), were tested in laboratory in order to select the most effective fungitoxicants against the pathogen under study .

1. RADIAL GROWTH OF FUNGAL COLONY :

The growth of fungal colonies was recorded in order to examine the efficacy of fungicides against the pathogen according to the technique described under "Material & Method". Fungitoxicants, which supported development of pathogen tested, were considered ineffective. The growth of fungus on various fungitoxicants, was measured and the average diameter of fungal colony on each fungitoxicant, was recorded along with control. Per cent inhibition over control was calculated and the results are summarised in Table XXXXIII and corresponding Figures 54 and 55.

TABLE - XXXXIII

Effect of different fungitoxicants on the radial growth of pathogen, *Alternaria alternata* (Fries.), Keissler, causing leaf Spot of Dolichos bean (*Dolichos lablab*, L.)

S. No.	Fungitoxicants	Dose (%)	Average Diameter of fungal colony (in mm.)	Percent inhibition over control
1.	Agrosan G.N.	0.2	0.00	100.00
2.	Aureofungin	0.1	0.00	100.00
3.	Bavistin	0.1	16.30	84.79

4.	Benlate	0.1	45.80	51.84
5.	Blitox-50	0.3	72.50	23.14
6.	Brassicol	0.2	57.20	39.52
7.	Captan	0.2	0.00	100.00
8.	Calixin	0.3	80.70	14.60
9.	Ceresan (Dry)	0.2	0.00	100.00
10.	Dichlone	0.2	9.15	90.87
11.	Dithane M-45	0.2	3.95	96.48
12.	Dithane Z-78	0.2	11.68	87.91
13.	Duter	0.1	0.00	100.00
14.	Emisan-6	0.2	0.00	100.00
15.	Ferbam	0.2	22.09	76.80
16.	Foltaf 80-W	0.2	0.00	100.00
17.	Hexaferb	0.2	13.90	85.72
18.	Karathane	0.2	0.00	100.00
19.	Kavach	0.3	0.00	100.00
20.	Pancotine	0.1	0.00	100.00
21.	Ridomil	0.1	52.50	44.12
22.	Spergon	0.2	17.55	81.60
23.	Suflex	0.3	82.70	11.88
24.	Thiram	0.2	0.00	100.00
25.	Vitavax	0.1	0.00	100.00
26.	Ziram	0.2	18.90	80.87
27.	Control	—	94.20	—

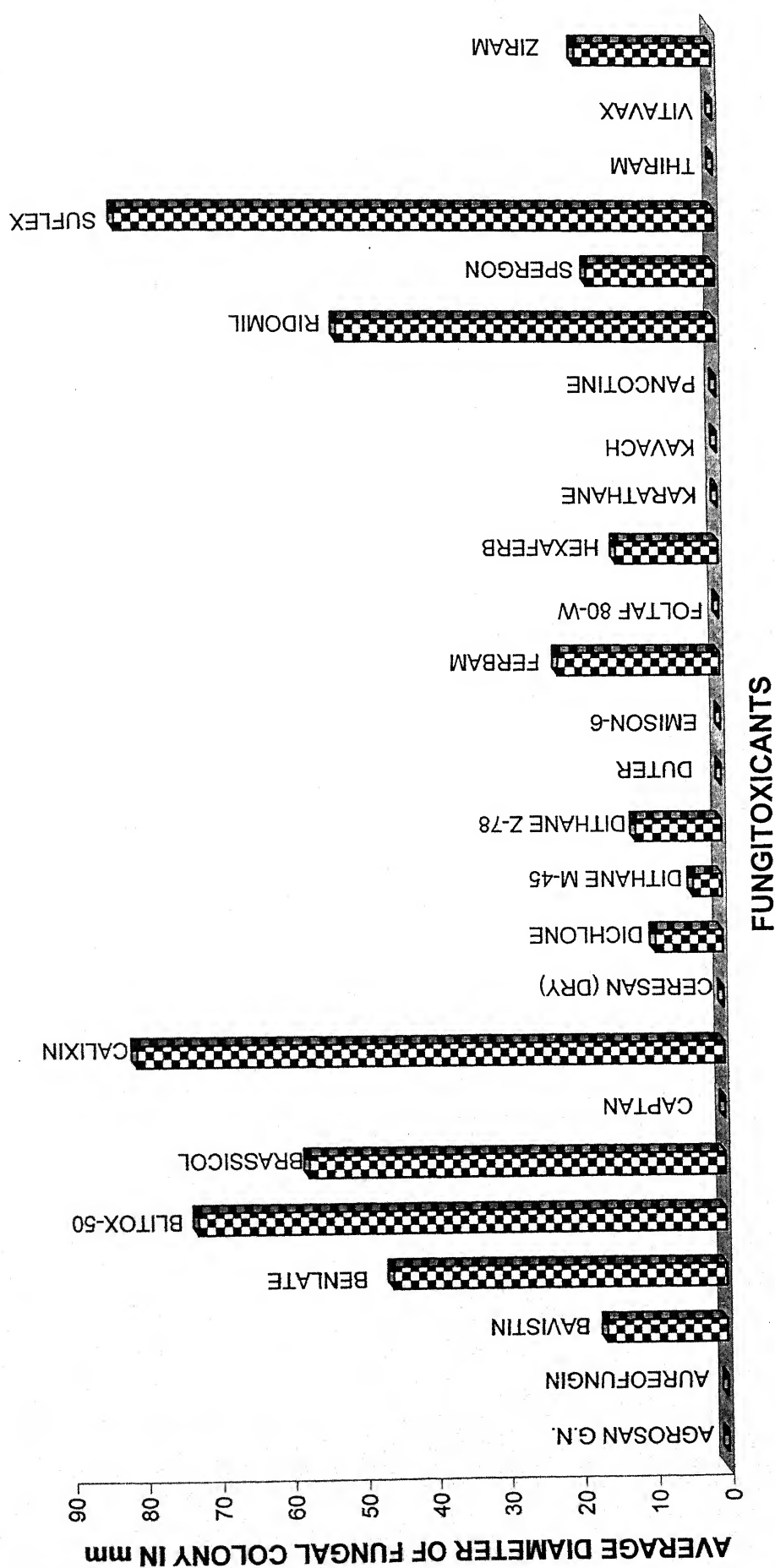
(—) = Denotes Absence.

FIGURE -54

Effect of different fungitoxicants on the radial growth of pathogen, *Alternaria alternata* (Fries.), Keissler, causing leaf spot disease of Dolichos bean (*Dolichos lablab*, L.).

1. Agrosan G.N.
2. Aureofungin.
3. Bavistin.
4. Benlate.
5. Blitox-50.
6. Brassicol.
7. Captan.
8. Calixin.
9. Ceresan (Dry).
10. Dichlone.
11. Dithane M-45.
12. Dithane Z-78.
13. Duter.
14. Emisan-6.
15. Ferbam.
16. Foltaf 80-W.
17. Hexaferb.
18. Karathane.
19. Kavach.
20. Pancotine.
21. Ridomil.
22. Spergon.
23. Suflex.
24. Thiram.
25. Vitavax.
26. Ziram.

FIGURE-54



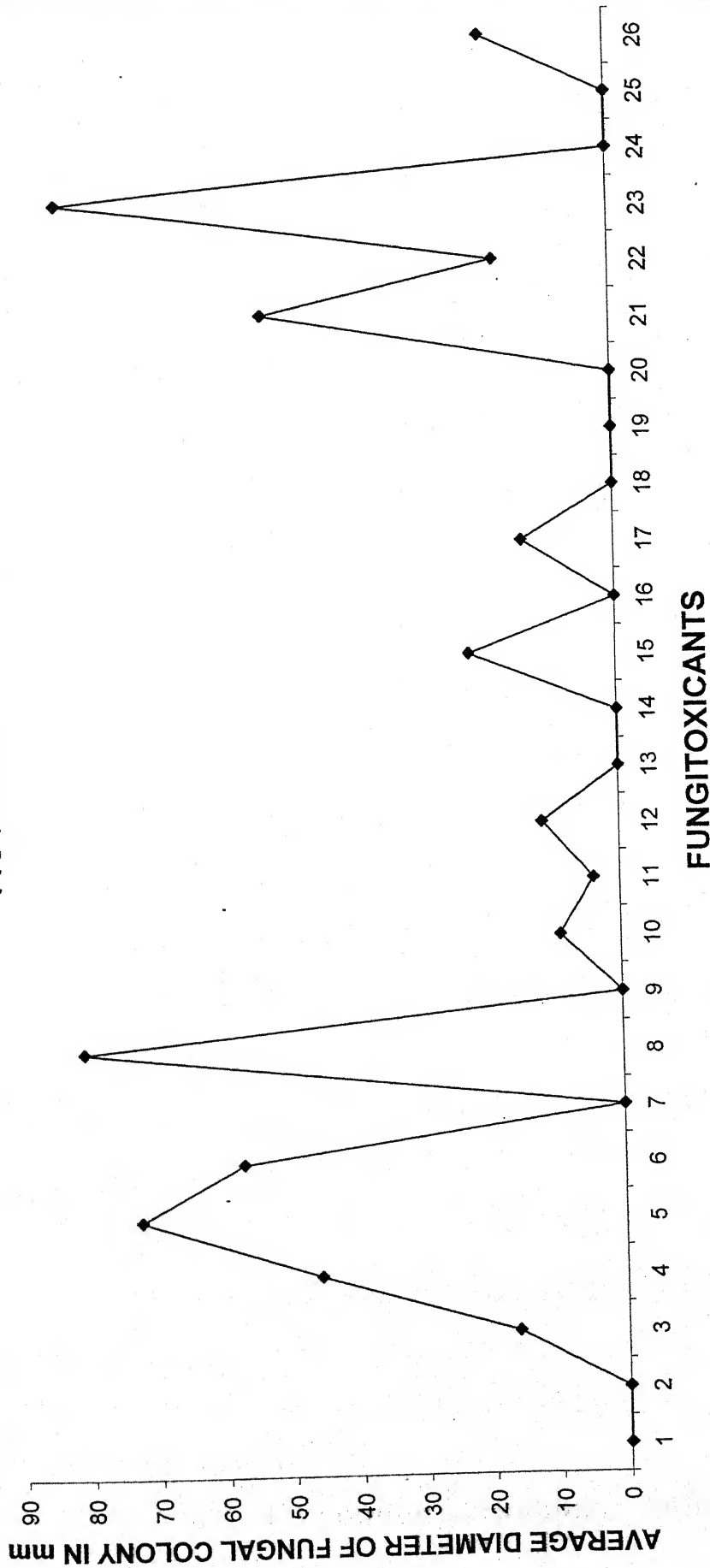
INHIBITORY EFFECT OF DIFFERENT FUNGITOXICANTS ON RADIAL GROWTH OF PATHOGEN *Alternaria alternata* (Fries.), Keissler, CAUSING LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.)

FIGURE - 55

Inhibitory effect of different Fungitoxicants in the radial growth of pathogen, *Alternaria alternata* (Fries.), Keissler causing Leaf spot Disease of Dolichos bean (*Dolichos lablab*, L.).

1. Agrosan G.N.
2. Aureofungin
3. Bavistin
4. Benlate
5. Blitox-50
6. Brassicol
7. Captan
8. Calixin
9. Ceresan (Dry)
10. Dichlone
11. Dithane M-45
12. Dithane Z-78
13. Duter
14. Emisan-6
15. Ferbam
16. Foltaf 80-W
17. Hexaferb
18. Karathane
19. Kavach
20. Pancotine
21. Ridomil
22. Spergon
23. Fuflex
24. Thiram
25. Vitavax
26. Ziram

FIGURE-55



INHIBITORY EFFECT OF DIFFERENT FUNGITOXICANTS ON RADIAL GROWTH OF PATHOGEN
Alternaria alternata (Fries.), Keissler, CAUSING LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos*
lablab, L.)

It is evident from the data presented in Table XXXXIII and Figures 54 and 55 that all the fungitoxicants, were significant better in performance in inhibiting the growth of the pathogen in comparison to control. Out of 25 fungicides an anitbiotic (Aureofungin) examined, Agrosan G.N., Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax proved to be most effective as they inhibited the growth of fungus. Other fungitoxicants viz., Dithane M-45 and Dichlone, were found effective in checking the growth of fungus up to 96.48 per cent and 90.87 per cent respectively over control. The remaining fungitoxicants viz., Suflex, Calixin, Blitox-50, Brassicol, Ridomil, Benlate, Ferbam, Ziram, Spergon, Bavistin, Hexaferb and Dithane Z-78 were found partially effective in checking the growth of fungus upto 11.88 per cent, 14.60 per cent, 23.14 per cent, 39.52 per cent, 44.12 per cent, 51.84 per cent, 76.80 per cent, 80.37 per cent 81.60 per cent, 84.79 per cent, 85.72 per cent and 87.91 per cent respectively over control.

2-HYPHAL DRY WEIGHT :

The efficacy of fungitoxicants was tested by the technique described under "Material and Method". The result of dry weight in different concentrations of various fungitoxicants, viz., 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent, and 0.60 per cent, was recorded alongwith control. Per cent inhibition over control was calculated and results are summarised in table XXXXIV and corresponding Figures 56, 57 and 58.

It is evident from the results summarised in Table XXXXIV and Figures 56, 57 and 58 that all the fungitoxicants used in different concentrations viz, 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent, and 0.60 per cent, were significantly better in performance in comparison to control. Out of 25 fungicides and antibiotic (Aureofungin) examined, Agrosan G.N.,

TABLE - XXXXIV

Efficacy of Fungitoxicants in various concentrations against pathogen *Alternaria alternata* (Fries.), Keissler on growth.

S. No.	Fungitoxicants	Concentrations of Fungitoxicants used											
		0.05		0.10		0.15		0.20		0.40		0.60	
		Average dry weight of Fungal mycelium (mg.)	Percent inhibition over control	Average dry weight of Fungal mycelium (mg.)	Percent inhibition over control	Average dry weight of Fungal mycelium (mg.)	Percent inhibition over control	Average dry weight of Fungal mycelium (mg.)	Percent inhibition over control	Average dry weight of Fungal mycelium (mg.)	Percent inhibition over control	Average dry weight of Fungal mycelium (mg.)	Percent inhibition over control
1.	Agrosan G.N.	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
2.	Aureofungin	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
3.	Bavistin	72.90	25.40	66.38	31.71	62.20	35.80	60.50	38.88	56.20	43.07	52.77	45.65
4.	Benlate	42.30	56.84	37.24	61.89	33.15	65.79	30.25	68.97	28.60	70.27	24.8	74.88
5.	Blitox - 50	16.35	82.86	13.68	86.34	11.20	88.76	12.30	87.29	10.55	89.34	7.59	92.00
6.	Brassicol	27.92	71.41	22.29	77.16	20.36	79.62	17.95	81.73	12.69	87.04	12.40	87.36
7.	Captan	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
8.	Calixin	13.70	85.70	10.70	88.60	10.90	88.98	10.25	70.48	8.35	91.20	4.30	95.69
9.	Ceresan (Dry)	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
10.	Dichlone	82.60	14.71	77.30	20.77	78.54	19.50	70.36	27.61	65.06	33.08	63.27	35.00
11.	Dithane M-45	94.50	2.95	84.19	13.18	82.44	17.24	78.25	19.29	78.90	19.05	68.90	29.47
12.	Dithane Z-78	77.40	20.67	70.50	27.56	66.65	31.43	66.28	31.72	68.74	29.27	59.84	38.83
13.	Duter	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
14.	Emisan-6	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00

15.	Ferbam	48.60	50.20	44.20	54.32	40.25	58.58	38.59	60.91	38.62	60.37	33.67	65.25
16.	Foltaf 80-W	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
17.	Hexaferb	72.29	25.83	67.03	31.03	64.90	33.13	60.0	38.37	60.30	37.87	58.10	40.22
18.	Karathane	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
19.	Kavach	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
20.	Pancotine	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
21.	Ridomil	38.70	60.29	29.92	69.22	27.82	71.27	28.81	70.15	28.75	70.11	23.30	75.82
22.	Spergon	67.0	31.16	58.20	40.32	53.28	45.38	54.70	43.41	49.70	53.80	40.84	59.47
23.	Suflex	10.97	88.91	7.54	92.44	6.50	93.41	8.43	91.33	5.20	94.85	4.28	95.79
24.	Thiram	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
25.	Vitavax	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
26.	Ziram	62.50	35.70	52.90	54.62	53.0	51.34	47.70	50.92	42.50	56.69	39.35	59.71
27.	Control	97.30	--	97.30	-	97.10	-	97.10	-	97.20	-	97.30	-

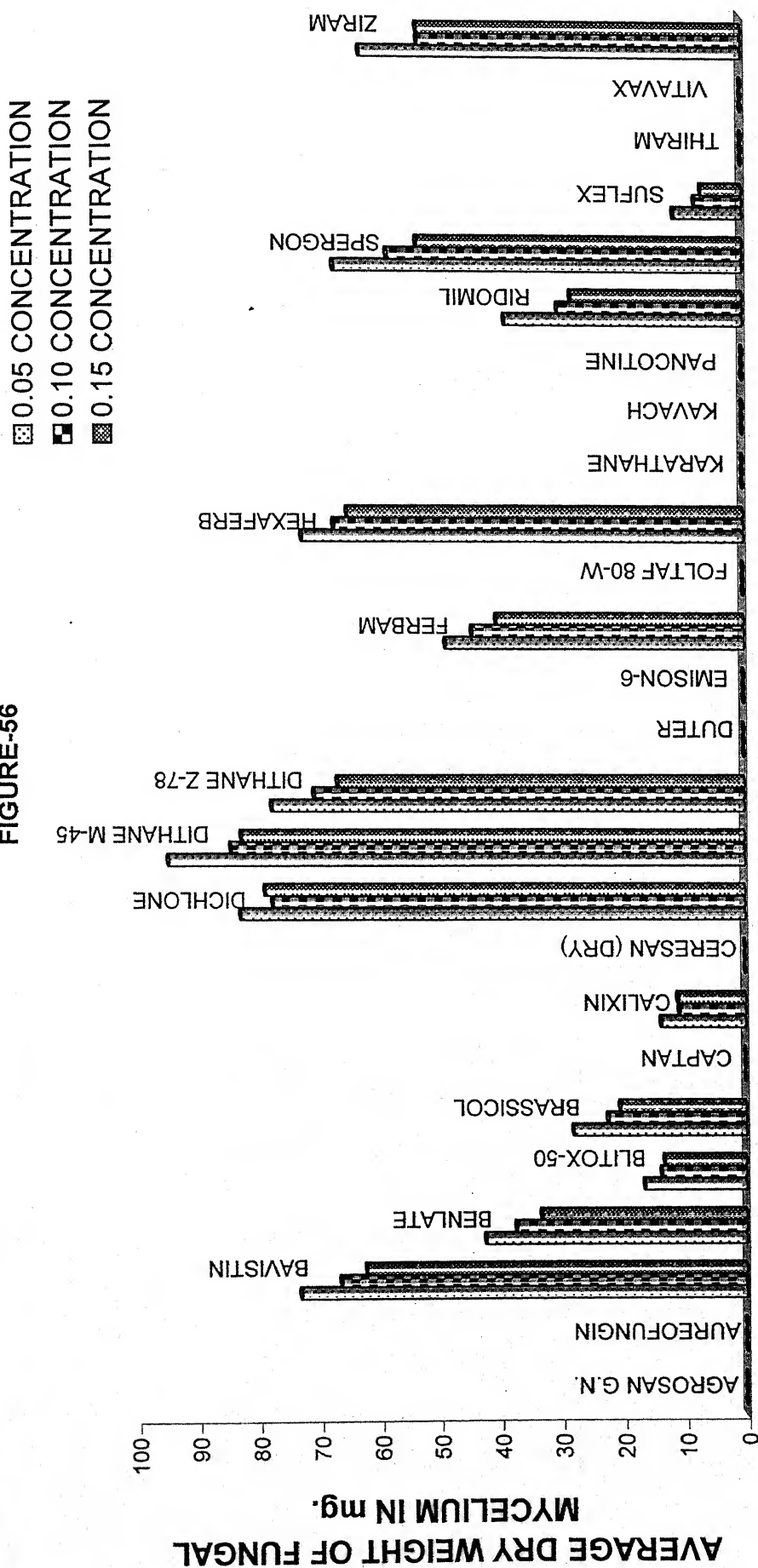
(-) = Absence of Infection over control.

FIGURE -56

Efficacy of Fungitoxicants in various concentrations against pathogen *Alternaria alternata* (Fries.), Keissler on growth.

1. Agrosan G.N.
2. Aureofungin.
3. Bavistin.
4. Benlate.
5. Blitox-50.
6. Brassicol.
7. Captan.
8. Calixin.
9. Ceresan (Dry).
10. Dichlone.
11. Dithane M-45.
12. Dithane Z-78.
13. Duter.
14. Emisan-6.
15. Ferbam.
16. Foltaf 80-W.
17. Hexaferb.
18. Karathane.
19. Kavach.
20. Pancotine.
21. Ridomil.
22. Spergon.
23. Suflex.
24. Thiram.
25. Vitavax.
26. Ziram.

FIGURE-56



DIFFERENT CONCENTRATIONS OF FUNGITOXICANTS

EFFICACY OF FUNGITOXICANTS IN VARIOUS CONCENTRATIONS AGAINST PATHOGEN *Alternaria alternata* (Fries.), Keissler, ON GROWTH.

FIGURE -57

Efficacy of Fungitoxicants in various concentrations against pathogen *Alternaria alternata* (Fries.), Keissler on growth.

1. Agrosan G.N.
2. Aureofungin.
3. Bavistin.
4. Benlate.
5. Blitox-50.
6. Brassicol.
7. Captan.
8. Calixin.
9. Ceresan (Dry).
10. Dichlone.
11. Dithane M-45.
12. Dithane Z-78.
13. Duter.
14. Emisan-6.
15. Ferbam.
16. Foltaf 80-W.
17. Hexaferb.
18. Karathane.
19. Kavach.
20. Pancotine.
21. Ridomil.
22. Spergon.
23. Suflex.
24. Thiram.
25. Vitavax.
26. Ziram.

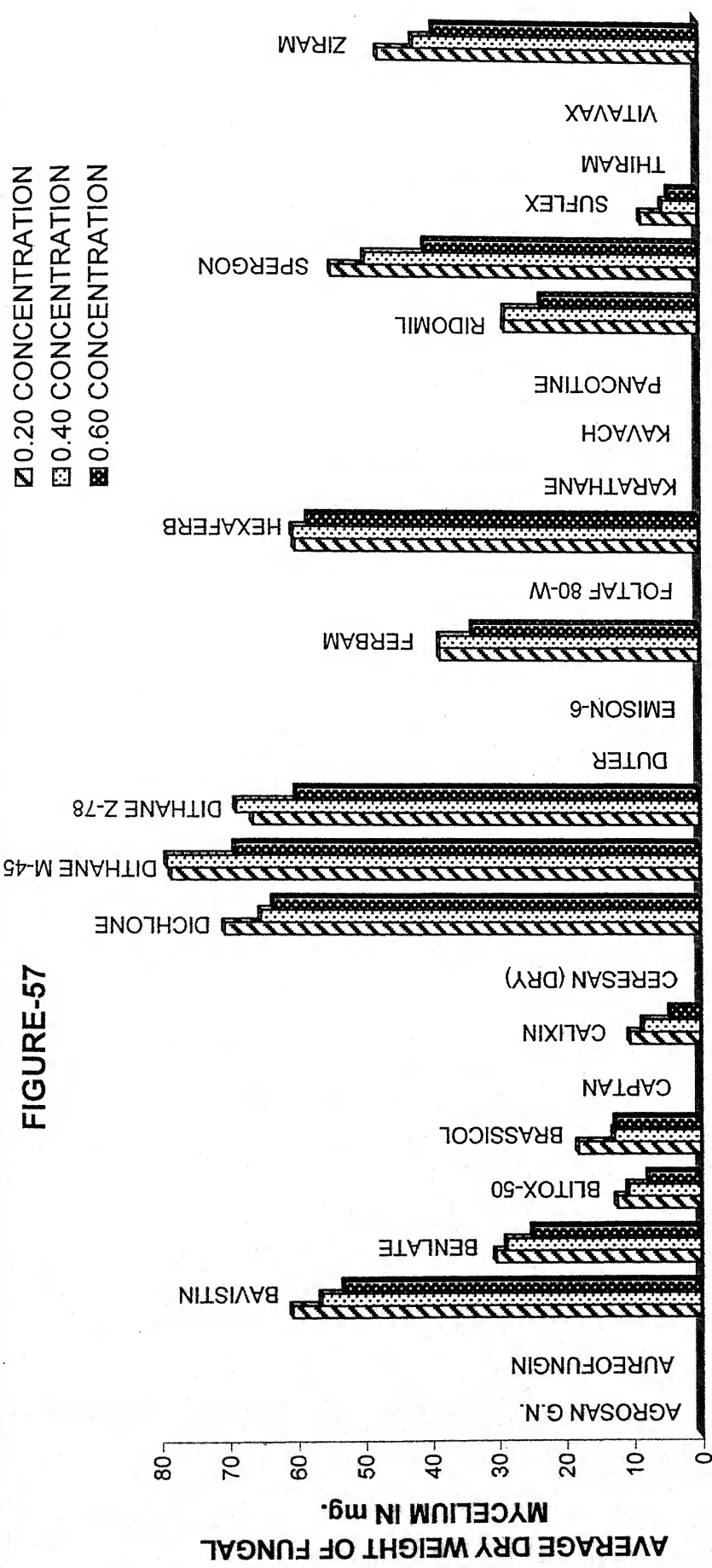
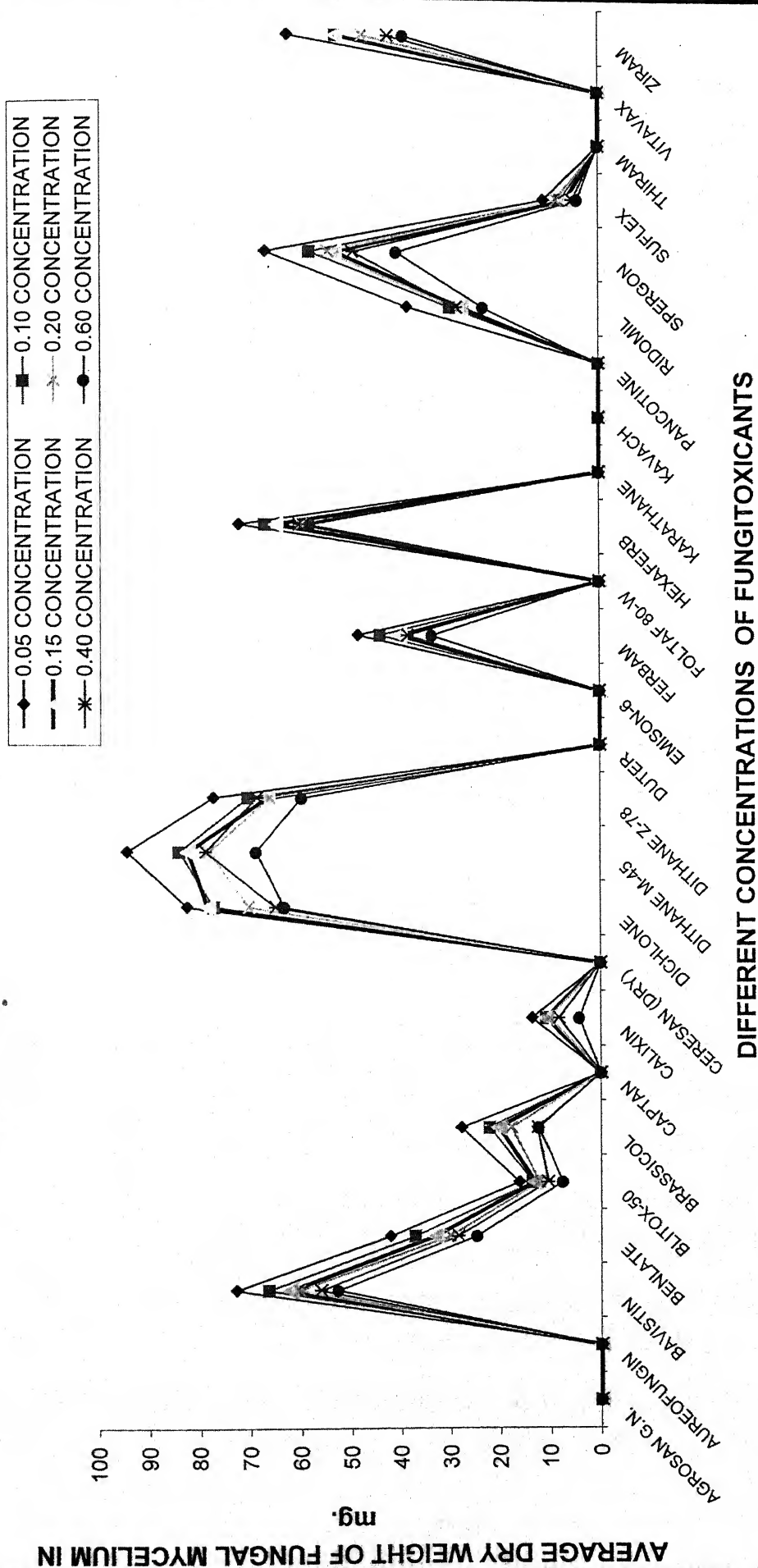


FIGURE -58

Efficacy of Fungitoxicants in various concentrations against pathogen *Alternaria alternata* (Fries.), Keissler on growth.

1. Agrosan G.N.
2. Aureofungin.
3. Bavistin.
4. Benlate.
5. Blitox-50.
6. Brassicol.
7. Captan.
8. Calixin.
9. Ceresan (Dry).
10. Dichlone.
11. Dithane M-45.
12. Dithane Z-78.
13. Duter.
14. Emisan-6.
15. Ferbam.
16. Foltaf 80-W.
17. Hexaferb.
18. Karathane.
19. Kavach.
20. Pancotine.
21. Ridomil.
22. Spergon.
23. Suflex.
24. Thiram.
25. Vitavax.
26. Ziram.

FIGURE -58.



DIFFERENT CONCENTRATIONS OF FUNGITOXICANTS

EFFICACY OF FUNGITOXICANTS IN VARIOUS CONCENTRATIONS AGAINST PATHOGEN *Alternaria alternata* (Fries.), Keissler, ON GROWTH.

Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax proved to be most effective as they inhibited the growth of fungus. The other fungitoxicants viz; Calixin and Suflex, were found effective in arresting the growth of fungus upto 85.70 per cent at 0.05 per cent, 88.60 per cent at 0.10 per cent, 88.98 per cent at 0.15 per cent, 70.48 per cent at 0.20 per cent, 91.20 per cent at 0.40 per cent and 95.69 per cent at 0.60 per cent and 88.91 per cent, at 0.05 per cent, 92.44 per cent at 0.10 per cent, 93.41 per cent in 0.15 per cent, 91.33 per cent in 0.20 per cent, 94.85 per cent in 0.40 per cent and 95.79 per cent in 0.60 per cent concentrations respectively, the per cent inhibition over control. The remaining fungitoxicants viz., Bavistin, Benlate, Blitox-50, Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Hexaferb, Ridomil, Sprrgon and Ziram, were found partially effective in checking the growth of fungus in various concentrations of fungitoxicants used. The significant variations in inhibition in hyphal dry weight with respect to the treatment of differtent fungitoxicants as well as their different concentrations.

3. LESION DEVELOPMENT TEST :

A dosage response was studied in different concentrations viz., 0.05 per cent , 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent and 0.60 per cent of fungitoxicants under study and pathogenic effects of pathogen, *Alternaria alternata* (Fries.), Keissler according to the technique described under "Material and Method". The results of pathogenic effect viz., light yellow lesion (LY), Yellowish brown lesion (YB), Light brown lesion(LB), Extensive brown lesion (EB), and Phytotoxic effect (PE), were observed in different concentrations of fungitoxicants alongwith control. The observations regarding the type of pathogenic effects produced are summarised in Table XXXXV and corresponding Figure 59.

TABLE - XXXXV

Efficacy of fungitoxicants in various concentrations against pathogen *Alternaria alternata* (Fries.), Keissler, causing leaf Spot of Dolichos bean (*Dolichos lablab* L.)

S. No.	Fungi-toxicants	Concentrations of Fungicides											
		0.05		0.10		0.15		0.20		0.25		0.30	
		Grade	Pathogenic Reaction	Grade	Pathogenic Reaction	Grade	Pathogenic Reaction	Grade	Pathogenic Reaction	Grade	Pathogenic Reaction	Grade	Pathogenic Reaction
1.	Agrosan G.N.	0	-	0	-	0	-	0	-	0	-	0	-
2.	Aureofungin	0	-	0	-	0	-	0	-	0	-	0	-
3.	Bavistin	4	EB	3	LB	4	EB	4	EB	4	EB	4	EB
4.	Benlate	2	YB	2	YB	2	YB	2	YB	2	YB	2	LY
5.	Blitox-50	2	YB	2	YB	2	YB	2	YB	2	YB	2	YB
6.	Brassiccol	1	LY	1	LY	1	LY	1	LY	1	LY	-	-
7.	Captan	0	-	0	-	0	-	0	-	0	-	0	-
8.	Calixin	1	LY	1	LY	1	LY	1	LY	1	LY	1	LY
9.	Ceresan (Dry)	0	-	0	-	0	-	0	-	0	-	0	-
10.	Dichlone	4	EB	4	EB	4	EB	4	EB	3	LB	3	LB
11.	Dithane M-45	5	PE	5	PE	5	PE	5	PE	5	PE	5	PE
12.	Dithane Z-78	4	EB	4	EB	4	EB	4	EB	4	EB	4	EB
13.	Duter	0	-	0	-	0	-	0	-	0	-	0	-
14.	Emisan-6	0	-	0	-	0	-	0	-	0	-	0	-
15.	Ferbam	4	EB	3	LB	3	LB	3	LB	3	LB	3	LB
16.	Foltaf 80-W	0	-	0	-	0	-	0	-	0	-	0	-
17.	Hexaferb	3	LB	2	YB	2	YB	2	YB	2	YB	2	YB

18.	Karathane	0	-	0	-	0	-	0	-	0	-	0	-
19.	Kavach	0	-	0	-	0	-	0	-	0	-	0	-
20.	Pancotine	0	-	0	-	0	-	0	-	0	-	0	-
21.	Ridomil	4	EB	3	LB	3	LB	3	LB	3	LB	3	LB
22.	Sperguson	4	EB	3	LB	3	LB	3	LB	3	LB	3	LB
23.	Suflex	1	LY	1	LY	1	LY	1	LY	1	LY	1	LY
24.	Thiram	0	-	0	-	0	-	0	-	0	-	0	-
25.	Vitavax	0	-	0	-	0	-	0	-	0	-	0	-
26.	Ziram	4	EB	4	EB	4	EB	4	EB	4	EB	4	EB
27.	Control	0	-	0	-	0	-	0	-	0	-	0	-

(-) = Denotes Absence, (0) = Denotes Nil, (LY) = Denotes Light Yellow lesions.
 (YB) = Denotes yellow brown lesions. (LB) = Denotes Light Brown Lesions
 (EB) = Denotes Extensive brown lesions. (PE) = Denotes Phytotoxic effects.

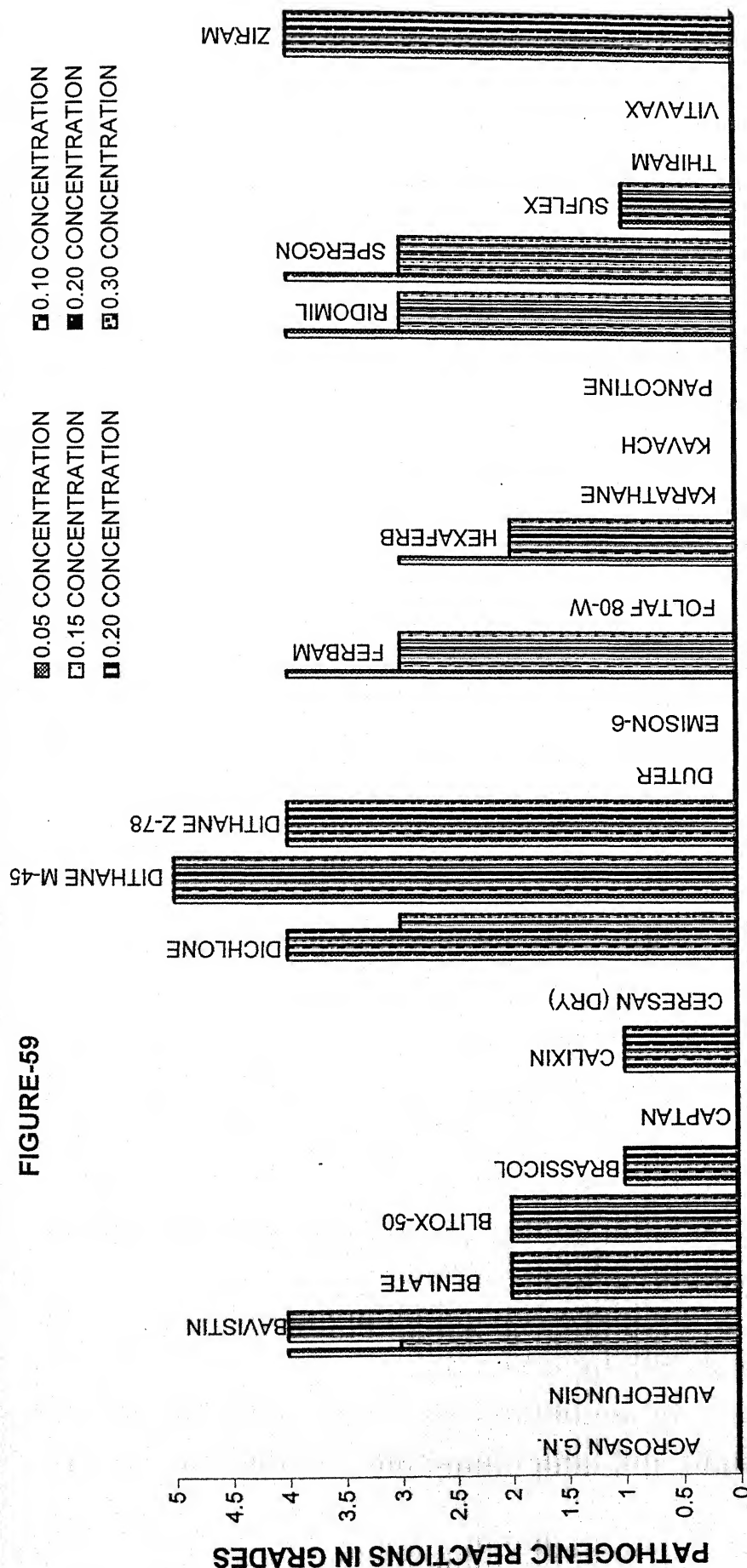
It is evident from the data summarised in Table XXXXV and Figure 59 that all the fungitoxicants, used in different concentrations of 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent, and 0.60 per cent, were better in performance in comparison to control. Out of the 24 fungicides and an antibiotic (Aureofungin) tested, Agrosan G.N., Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine. Thiram and Vitavax to be most effective as did not produced any pathogenic effects. Other fungitoxicants, viz., Brassicol, Calixin, and Suflex were found effective in producing the light yellow lesions (LY), only in different concentrations of fungitoxicants and proved better in comparison to the remaining fungitoxicants tested except Brassicol, in 0.30 per cent, the higher concentration, where no pathogenic effects were showed. The phytotoxic effects (PE), were recorded in the treatment with Dithane M-45, while extensive brown spots (EB), were observed in the treatment with Bavistin, Dithane Z-78 and Ziram in different concentrations except light brown lesions (LB) in 0.10 per cent concentration in

FIGURE -59

Efficacy of fungitoxicants in various concentrations against pathogen *Alternaria alternata* (Fries.), Keissler, causing leaf spot disease of Dolichos bean (*Dolichos lablab*. L.).

1. Agrosan G.N.
2. Aureofungin.
3. Bavistin.
4. Benlate.
5. Blitox-50.
6. Brassicol.
7. Captan.
8. Calixin.
9. Ceresan (Dry).
10. Dichlone.
11. Dithane M-45.
12. Dithane Z-78.
13. Duter.
14. Emisan-6.
15. Ferbam.
16. Foltaf 80-W.
17. Hexaferb.
18. Karathane.
19. Kavach.
20. Pancotine.
21. Ridomil.
22. Spergon.
23. Suflex.
24. Thiram.
25. Vitavax.
26. Ziram.

FIGURE-59



DIFFERENT CONCENTRATIONS OF FUNGITOXICANTS

EFFICACY OF FUNGITOXICANTS IN VARIOUS CONCENTRATIONS AGAINST PATHOGEN *Alternaria alternata* (Fries.), Keissler, CAUSING LEAF SPOT DISEASE OF DOLICHOS BEAN (*Dolichos lablab*, L.)

(0) DENOTES NIL (1) DENOTES LIGHT YELLOW LESIONS (2) DENOTES YELLOWISH BROWN LESIONS (3) DENOTES LIGHT BROWN LESIONS (4) DENOTES EXTENSIVE BROWN LESIONS AND (5) DENOTES PHYTOTOXIC EFFECTS

the treatment with Bavistin. The fungitoxicants viz; Dichlone, Ferbam, Ridomil and Spergon produced firstly the extensive brown spots (EB) in lower concentration of 0.05 per cent and thereafter in higher concentrations i. e. 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.25 per cent, 0.30 per cent decreased the intensity of pathogenic effects exhibiting light brown lesions (LB), Benlate. Blitox-50 and Hexaferb shown yellowish brown lesions (YB) in different concentrations except Benlate and Hexaferb, which exhibited light yellow lesions (LY) in higher concentration of 0.30 per cent and lower concentration of 0.05 per cent the results showed that as the concentrations increased, the phytotoxic effects were found decreased.

4. SPORE GERMINATION TEST :

The effect of fungitoxicants in their different concentrations viz., 0.0002 per cent, 0.0004 per cent and 0.0006 per cent on germination, number of germ tubes and their length due to effect of the pathogen, *Alternaria alternata* (Fries.), Keissler according to the procedure described under "Material and Method". The results on the effect of fungitoxicants on spore germination, number of germ tubes and their length produced by spores are presented in Table XXXXVI.

It is clear from the Table XXXXVI, that all the fungitoxicants under study in their different concentrations viz., 0.0002 per cent, 0.0004 per cent and 0.0006 per cent were better in performance in comparison to control. Out of the 25 fungicides and an antibiotic (Aureofungin) tested, Agrosan G.N., Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80-W, Karathane, Kavach, Pancotine Thiram and Vitavax proved to be most effective as spores failed to germinate. The other fungitoxicants viz., Dithane M-45 and Vitavax were also found effective but produced minimum spore germination and proved better in comparison to other fungitoxicants tested. The maximum inhibition per cent in spore germination over control was found 79.46 per cent in 0.0002 per cent concentration, 81.41 per cent in 0.0004 per cent concentration and 84.05 per cent in 0.0006 per cent concentration. The remaining fungicides viz;

TABLE - XXXXVI

Efficacy of Fungitoxants in various concentrations against pathogen, *Alternaria alternata* (Fries.), Keissler, on germination of spore, number of germ tubes and length of germ tubes.

S. No.	Fungitoxants	Concentration of Fungitoxants use																	
		0.0002						0.0004						0.0006					
		Average spore germination	Percent inhibition over control	Number of germ tubes	Percent inhibition over control	Length of germ tube (mm.)	Percent inhibition over control	Average spore germination	Percent inhibition over control	Number of germ tubes	Percent inhibition over control	Length of germ tube (mm.)	Percent inhibition over control	Average spore germination	Percent inhibition over control	Number of germ tubes	Percent inhibition over control	Length of germ tube (mm.)	Percent inhibition over control
1.	Agrosan G.N.	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
2.	Aureofungin	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
3.	Bavistin	34	62.46	10	80.25	0.005	53.00	33	64.24	18	64.25	0.007	68.28	26	71.37	6	87.91	0.006	63.73
4.	Benlate	57	37.83	28	48.31	0.012	35.72	57	35.65	23	72.12	0.017	44.86	52	46.70	28	54.28	0.006	77.37
5.	Blitox - 50	75	20.83	36	37.40	0.017	24.06	64	27.78	27	68.78	0.016	36.76	55	40.52	27	50.77	0.013	45.35
6.	Brassicol	72	23.94	32	41.07	0.018	30.42	62	30.00	28	37.83	0.015	36.86	58	39.26	27	52.53	0.012	45.48
7.	Captan	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
8.	Calixin	60	18.35	37	32.24	0.017	24.82	61	31.22	30	41.60	0.014	34.74	60	36.17	29	54.54	0.013	40.70
9.	Ceresan (Dry)	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
10.	Dichlone	27	72.06	11	78.37	0.008	59.05	19	77.87	9	86.89	0.008	65.30	18	81.81	21	91.32	0.007	72.62
11.	Dithane M-45	18	79.46	6	82.04	0.006	68.72	14	81.41	5	53.23	0.008	65.10	14	84.05	2	89.27	0.012	54.64

12.	Dithane Z-78	29	68.47	12	78.18	0.009	59.09	29	68.47	11	80.00	0.008	63.63	28	69.56	3	94.54	0.006	72.72
13.	Duter	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
14.	Emisan-6	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
15.	Ferbam	50	45.06	28	51.48	0.013	35.72	47	48.78	20	62.16	0.012	57.00	47	48.92	16	71.93	0.008	63.65
16.	Foltaf 80-W	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
17.	Hexaferb	40	56.78	16	73.81	0.012	34.67	30	66.96	14	71.50	0.008	73.30	23	76.40	11	80.60	0.009	78.19
18.	Karathane	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
19.	Kavach	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
20.	Pancotine	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
21.	Ridomil	63	31.28	30	46.52	0.015	32.48	57	35.45	25	43.29	0.013	45.88	50	46.70	25	56.15	0.010	54.55
22.	Spergon	45	53.86	20	64.38	0.013	34.70	43	51.11	18	67.82	0.007	74.28	35	64.79	18	68.43	0.008	63.62
23.	Suflex	78	15.67	27	35.81	0.017	33.82	76	16.55	28	43.29	0.014	45.90	68	29.88	28	47.28	0.004	45.45
24.	Thiram	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
25.	Vitavax	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00	0.00	100.00
26.	Ziram	47	50.54	20	62.40	0.018	25.39	40	55.55	17	66.03	0.012	55.00	27	72.35	14	75.45	0.009	78.17
27.	Control	94	—	55	—	0.020	—	90	—	54	—	0.025	—	93	—	57	—	0.023	—

Bavistin, Benlate, Blitox-50, Brassicol, Calixin, Dichlone, Dithane-Z-78, Ferbam, Hexaferb, Ridomil, Suflex, Spergon and Ziram, were found particularly effective in arresting the spore germination. The significant variations in inhibition over control were also recorded ranging from 15.75 per cent to 81.81 per cent in different concentrations of fungicides tested in respect of germination of seeds.

The number of germ tubes varied from 6-37, 5-30 and 2-29 in 0.0002, 0.0004 and 0.0006 per cent concentrations of fungitoxicants respectively. The minimum 6, 5 and 2 germtubes were produced in the treatment with Dithane in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations respectively, while the maximum number of germtubes 37, 30 as well as 29 in the treatment with Calixin in 0.0002, per cent, 0.0004 per cent and 0.0006 per cent, concentrations respectively. Other fungitoxicants viz; Bavistin, Benlate, Blitox-50, Brassicol, Dichlone, Dithane-Z-78, Ferbam, Hexaferb, Ridomil, Spergon Suflex and Ziram produced 10, 23, 36, 32, 11, 12, 28, 16, 30, 20, 28 and 20 in 0.0002 per cent, concentration, whereas 18, 23, 27, 28, 9, 11, 20, 14, 25, 18, 28 and 17 germtubes were found respectively in 0.0004 per cent concentration of Bavistin, Benlate, Blitox-50, Brassicol, Dichlone, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram. In 0.0006 per cent concentration of fungitoxicants viz., Bavistin, Benlate, Blitox 50, Brassicol, Dichlone, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex, and Ziram produced 6, 28, 27, 27, 3, 11, 25, 18, 28 and 14 germ tubes respectively. The inhibition percentage over control varied from 32.24 per cent to 82.04 per cent, 43.29 per cent to 86.89 per cent and 47.28 to 91.32 per cent in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentration of different fungitoxicants respectively.

As far as the length of germtube is concerned, it varied in size from 0.005 mm. to 0.018 mm., 0.007 mm. to 0.16 mm. and 0.006 mm. to 0.014 mm. per cent in 0.0002, 0.0004, 0.0006 per cent, concentrations of different fungitoxicants. The minimum germtube length measuring 0.005 mm., 0.007 mm., and 0.006 mm., were recorded in the treatment with Bavistin in 0.0002, 0.0004, and 0.0006 percent concentrations of fungitoxicant respectively, whereas the maximum

number of the germtube measuring length 0.017 mm. and 0.014 mm. and 0.013 mm. with the treatment with Calixin in 0.0002, 0.0004 and 0.0006 per cent concentrations of fungitoxicants respectively. In the treatment with other fungitoxicants viz, Benlate, Blitox-50, Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex, and Ziram produced germtubes measuring 0.012 mm., 0.017 mm., 0.018 mm., 0.008 mm., 0.006 mm., 0.009 mm., 0.013 mm., 0.012 mm., 0.015 mm., 0.013 mm., 0.017 mm. and 0.018 mm. in length respectively in 0.0002 per cent concentration, while germtubes measuring 0.015, 0.016, 0.015 mm., 0.008 mm., 0.008 mm., 0.008 mm., 0.012 mm., 0.008 mm., 0.013 mm., 0.007 mm., 0.014 mm. and 0.012 mm. were recorded in 0.0004 per cent concentration of Benlate, Blitox-50, Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram. Bavistin, Benlate, Blitox-50, Brassicol, Calixin, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram produced germtubes measuring length of 0.009 mm., 0.006 mm., 0.013 mm., 0.012 mm., 0.013 mm., 0.007 mm., 0.012 mm., 0.008 mm., 0.008 mm., 0.009 mm., 0.010 mm., 0.008 mm., 0.004 mm. and 0.009 mm., and 0.009 mm. respectively.

The inhibition percentage over control varied from 24.82 per cent to 68.72 per cent, 34.74 per cent to 74.28 per cent and 45.48 per cent to 78.19 per cent in 0.0002, 0.0004 and 0.0006 per cent concentrations of different fungitoxicants respectively.

The inhibition in all the three parameters in spore germination, germtube length and number of germtubes produced by the spores, were pronounced in higher concentration of fungitoxicants with significant differences between different fungitoxicants and their concentrations. Dichlone and Dithane M-45 caused appreciable degree of inhibition in spore germination, germ tube number and germ tube length. The germtube length was inhibited nearly 78.19 per cent at higher concentrations of these fungicides. At lower concentration Blitox-50 and Calixin, were not found to be effective but effective at higher concentrations of the inhibition of spore germination.

EFFECT OF FUNGITOXICANTS IN VITRO AGAINST *Alternaria alternata* (Fries.), Keissler, ON SEED GERMINATION AND SEED BORNE INFECTION :

The efficacy of selected seed dressing fungitoxicants viz., Agrosan G.N., Aureofungin (Antibiotic), Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine Thiram and Vitavax as seed dressers, which proved effective in laboratory. Screening was tested against *Alternaria alternata* (Fries.), Keissler on seed germination and seed borne infection in *vitro* according to the technique described under "Material and Method". The data on seed germination and seed borne infection were recorded as summarised in Table XXXXVII and corresponding Figure 60.

TABLE - XXXXVII

Effect of different seed dressing fungitoxicants on seed germination and seed borne infection of *Alternaria alternata* (Fries.), Keissler, in *vitro* in laboratory.

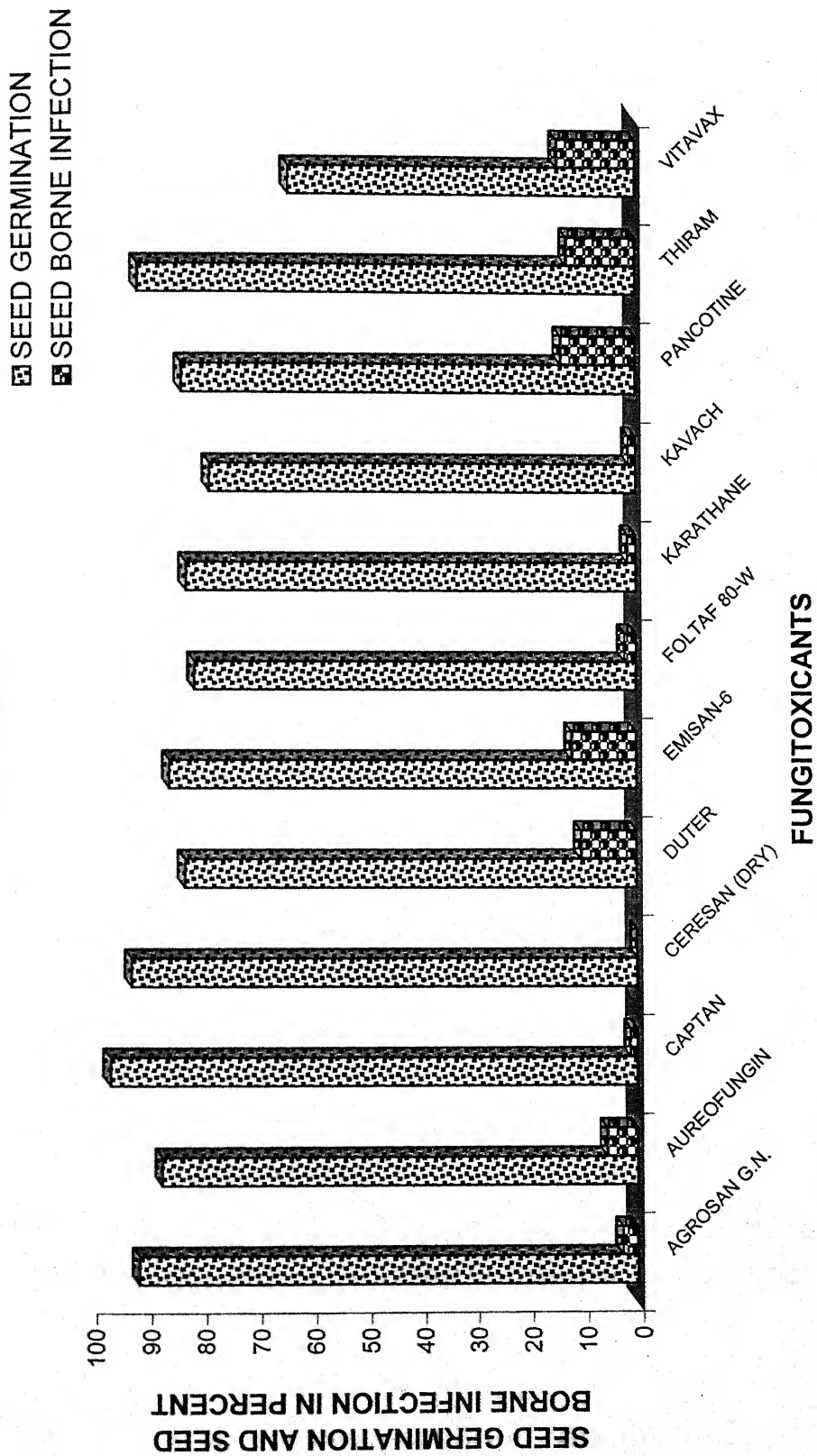
S. No.	Fungitoxicants	Dose (0/0)	Seed germination (0/0)	Percent increase in seed germination over control	Percent reduction in Seedling infection (0/0)	Percent reduction in seedling infection over control
1.	Agrosan G.N.	0.2	91.30	22.13	2.83	86.42
2.	Aureofungin	0.1	87.18	19.57	5.58	72.57
3.	Captan	0.2	96.49	14.87	1.29	88.03
4.	Ceresan (Dry)	0.2	92.65	29.25	0.00	100.00
5.	Duter	0.1	83.00	14.81	10.26	49.60
6.	Emisan-6	0.2	85.84	16.15	11.91	43.80
7.	Foltaf 80-W	0.2	81.27	9.22	2.20	86.85
8.	Karathane	0.2	82.79	24.42	1.65	93.79

FIGURE -60

Effect of different seed dressing fungitoxics on seed germination and seed borne infection of *Alternaria alternata* (Fries.), Keissler, in vitro in laboratory.

1. Agrosan G.N.
2. Aureofungin.
3. Captan.
4. Ceresan (Dry).
5. Duter.
6. Emisan-6.
7. Foltaf 80-W.
8. Karathane.
9. Kavach.
10. Pancotin.
11. Thiram.
12. Vitavax.

FIGURE-60



EFFECT OF SEED DRESSING FUNGITOXICANTS ON SEED GERMINATION AND SEED BORNE INFECTION OF *Alternaria alternata* (Fries.), Keissler, IN VITRO IN LABORATORY

9.	Kavach	0.2	78.53	22.78	1.34	91.25
10.	Pancotine	0.2	83.54	13.38	13.83	36.55
11.	Thiram	0.2	91.47	11.60	12.79	38.93
12.	Vitavax	0.2	64.28	13.90	14.55	43.78
13.	Control	—	74.00	—	24.82	—

It is clear from the Table XXXXVII and Figure 60, that maximum seed germination 96.49 per cent in Captan, followed by 92.65 per cent in Ceresan (Dry), 92.65 per cent in Thiram 91.30 per cent in Agrosan G.N., 87.18 per cent in Aureofungin (Antibiotic), 85.84 per cent in Emisan-6, 83.0 per cent in Duter, 83.54 per cent in Pancotine, 82.79 per cent in Karathane, 81.27 per cent in Foltaf-80-W 78.53 per cent in Kavach and 64.28 per cent in Vitavax. The lowest seed germination 64.28 per cent was recorded in Vitavax, followed by control, where the seeds were not treated with any fungitoxicants.

It was also recorded that seeds treated with Ceresan (Dry) had no infection. Further it was also recorded that amongst the various fungitoxicants tested the lowest infection 1.29 per cent, was found in treatment with Captan. The maximum 14.55 per cent infection was recorded in the seeds treated with Vitavax followed by Pancotine. Thiram, Emisan-6, Duter, Aureofungin (Antibiotic), Agrosan G.N., Foltaf-80-W, Karathane, Kavach and Captan ranging from 13.83 per cent, 12.79 per cent, 11.91 per cent, 10.26 per cent, 5.85 per cent, 2.83 per cent, 2.20 and 1.65 per cent, 1.34 per cent, 1.29 per cent respectively. In general all the fungitoxicants, examined were significant over control in reducing the seed borne infection.

EFFECT OF FUNGITOXICANTS ON SEED GERMINATION AND SEEDLING INFECTION AGAINST *Alternaria alternata* (Fries.), Keissler IN POT EXPERIMENTS :

The efficacy of selected fungitoxicants viz; Agrosan G.N., Aureofungin (Antibiotic), Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax as done in Standard Blotter Method,

was tested in pot experiments in order to detect out their ability in improving seed germination and reducing the incidence of seedling infection according to the technique described under "Material and Method". The data on seed germination and seed borne infection were recorded as summarised in table XXXXVIII and corresponding Figure 61.

TABLE - XXXXVIII

Effect of different seed dressing fungitoxicants on seed germination and seedling infection against *Alternaria alternata* (Fries.), Keissler, in Pot Experiments.

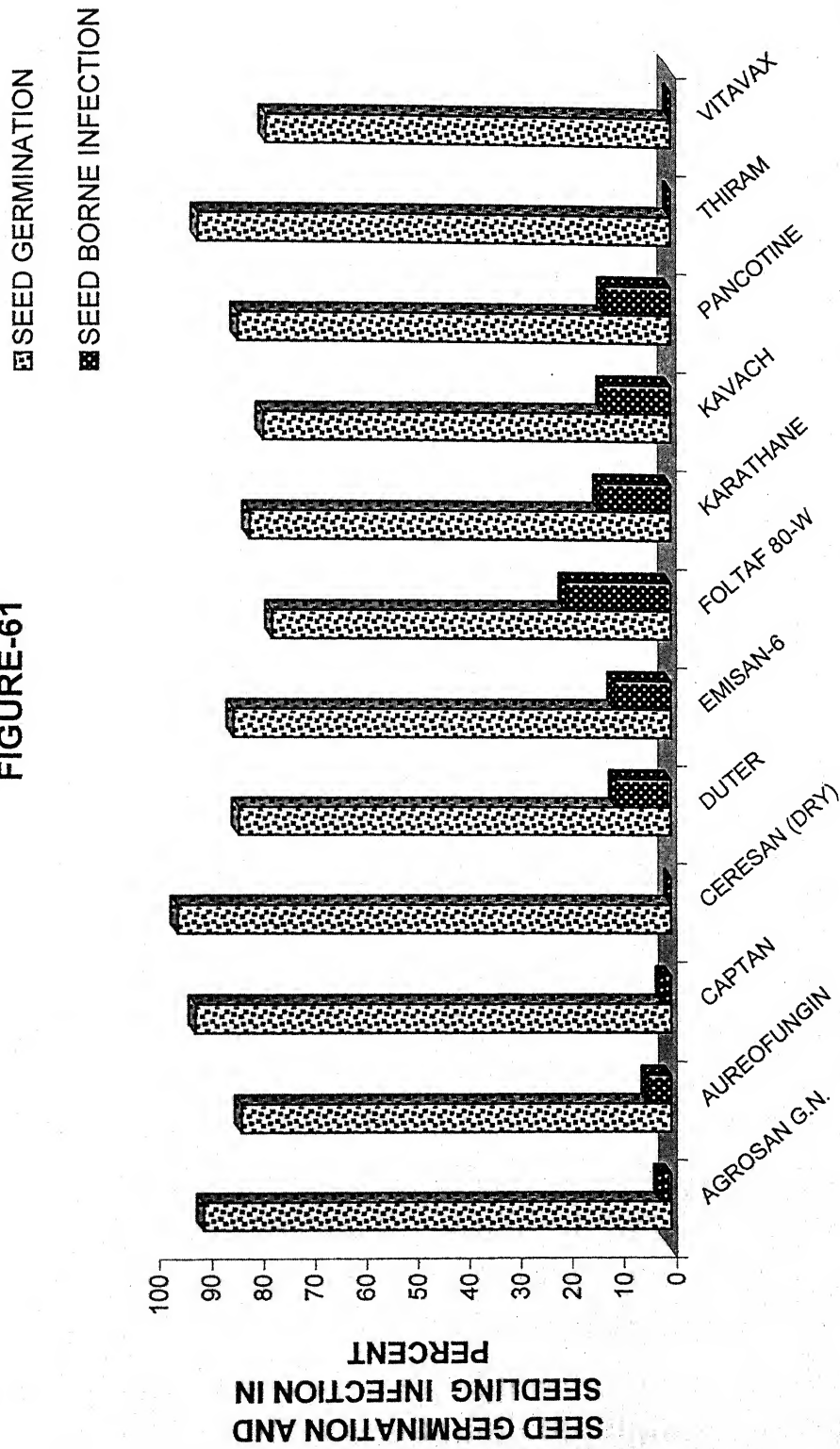
S. No.	Fungitoxicants	Dose (0/0)	Seed germination (0/0)	Percent increase in seed germination over control	Seedling infection (0/0)	Percent reduction in seedling infection over control
1.	Agrosan G.N.	0.2	90.45	21.89	1.83	91.54
2.	Aureofungin	0.1	83.19	12.24	4.39	78.97
3.	Captan	0.2	92.08	24.92	1.48	93.78
4.	Ceresan (Dry)	0.2	95.47	21.35	0.00	100.00
5.	Duter	0.1	83.75	12.97	10.57	54.03
6.	Emisan-6	0.2	84.78	14.20	10.89	48.65
7.	Foltaf 80-W	0.2	77.34	14.67	20.30	23.08
8.	Karathane	0.2	81.67	9.73	13.58	34.39
9.	Kavach	0.2	79.15	8.59	12.94	33.27
10.	Pancotine	0.2	83.98	12.59	12.76	37.82
11.	Thiram	0.2	91.57	23.82	0.00	100.00
12.	Vitavax	—	78.60	5.74	0.00	100.00
13.	Control	—	74.26	—	20.48	—

FIGURE -61

Effect of different seed dressing fungitoxicans on seed germination and seedling infection against *Alternaria alternata* (Fries.), Keissler, in pot experiments.

1. Agrosan G.N.
2. Aureofungin.
3. Captan.
4. Ceresan (Dry).
5. Duter.
6. Emisan-6.
7. Foltaf 80-W.
8. Karathane.
9. Kavach.
10. Pancotin.
11. Thiram.
12. Vitavax.

FIGURE-61



FUNGITOXICANTS

EFFECT OF SEED DRESSING FUNGITOXICANTS ON SEED GERMINATION AND SEEDLING INFECTION OF *Alternaria alternata* (Fries.), Keissler, IN POT EXPERIMENT.

It is obvious from the table XXXXVII and figure 60 that maximum germination 95.47 per cent was recorded in Ceresan (Dry), followed by 92.08 per cent in Captan, 91.57 per cent in Thiram, 90.45 per cent in Agrosan G.N., 84.78 per cent in Emisan-6, 83.75 per cent in Duter, 83.98 per cent in Pancotine, 83.79 per cent in Aureofungin (Antibiotic), 81.67 per cent in Karathane, 79.15 per cent in Kavach, 78.60 per cent in Vitavax, while poor germination 77.34 per cent in Foltaf-80w was recorded. Lowest seed germination 74.26 per cent was recorded in control, where the seeds were not treated with any fungitoxicants.

It was also observed that seed treatment with Ceresan (Dry), Thiram and Vitavax had no infection and highest infection 20.48 per cent was recorded in control. Captan and Agrosan G.N., were found superior in eliminating the seedling infection viz; 1.48 per cent and 1.83 per cent respectively. Other fungitoxicants viz., Aureofungin (Antibiotic), Duter, Emisan-6, Karathane, Kavach, Pancotine and Foltaf-80, were also found effective in reducing seedling infection i.e., 4.39 per cent, 10.75 per cent, 18.85 per cent, 13.58 per cent, 12.96 per cent, 12.94 per cent and 20.30 per cent respectively as compared to control. The maximum seedling infection 20.30 per cent was recorded in the treatment with Foltaf-80-W, followed by Karathane.

Seedling infection was significantly reduced in all the fungitoxicants tested over control, but was 100.0 per cent reduced in the seeds treated with Ceresan (Dry) Thiram and Vitavax followed by Captan and Agrosan G.N., were proved next reducing in seedling infection as they exhibited 93.78 per cent and 91.54 per cent reduction in seedling infection over control.

Thus in general the fungitoxicants belonging to heterocyclic nitrogen compound (Captan), Organomercury (Ceresan and Agrosan G.N.) compound groups and systemic (Vitavax) were most efficacious in comparison to the fungitoxicants of Dithiocarbamates, Benzene, Copper, Sulphur and Quinone compounds of systemic nature controlling the seed borne infection of *Alternaria alternata* (Fries.), Keissler associated with Dolichos bean seeds.

EVALUATION OF FUNGICIDES IN THE FIELD AS SPRAY :

Relative efficacy of eleven different fungitoxicants viz; Agrosan G.N., Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80-W, Karathane, Pancotine, Thiram and Vitavax as well as an antibiotic (Aureofungin), which were found completely or partially effective in bioassay test, were evaluated under pot and field conditions during the years 2001 and 2002 in order to select out suitable fungitoxicants for reducing the incidence of disease and boosting the seed yield. The method for the inoculation and spraying was followed as given in "Material and Method." The most susceptible variety, "Kalyanpur Type-1", was selected for the experiment and data on disease intensity and seed yield were taken as given in Table XXXXIX and XXXXX and Figures 62, 63, 64 and 65.

1. EVALUATION OF FUNGITOXICANTS IN POT EXPERIMENT :

It is evident from the Table XXXXIX and corresponding Figures 62 and 63 that all the fungitoxicants tested proved significantly effective in controlling the disease over control. On examining the relative efficacy of fungitoxicants, it was found that Captan, was proved best in controlling the disease during both the years 2001 and 2002 of trial and controlled the disease to the extent of 80.72 per cent and 81.49 per cent respectively. The next in effectiveness was Thiram, which controlled the disease up to 74.68 per cent and 78.15 per cent respectively in the years 2001 and 2002, followed by Ceresan dry, Agrosan G.N., Karathane, Kavach Duter, Aureofungin (Antibiotic), Emisan-6. Pancotine and Vitavax. Vitavax fungitoxicant, was found poorest amongst all the fungitoxicants tested in controlling the disease in both years 2001 and 2002. The highest disease intensity was recorded in control, where no fungitoxicants were employed.

In the years 2001 and 2002 of trial the disease, was controlled to the extent of 80.22 per cent and 81.49 per cent respectively. The next in effectiveness was Thiram, which controlled the disease up to 74.68 per cent and 78.15 per cent respectively in the year 2001 and 2002 followed by Ceresan (Dry), Agrosan G.N.; Karathane, Kavach, Aureofungin (Antibiotic), Pancotine, Emisan-6, Vitavax and

TABLE - XXXXIX

Effect of spraying of Fungitoxicants on the intensity of leaf Spot disease caused by *Alternaria alternata* (Fries.), Keissler, of *Dolichos lablab*, L.) in Pot Experiments.

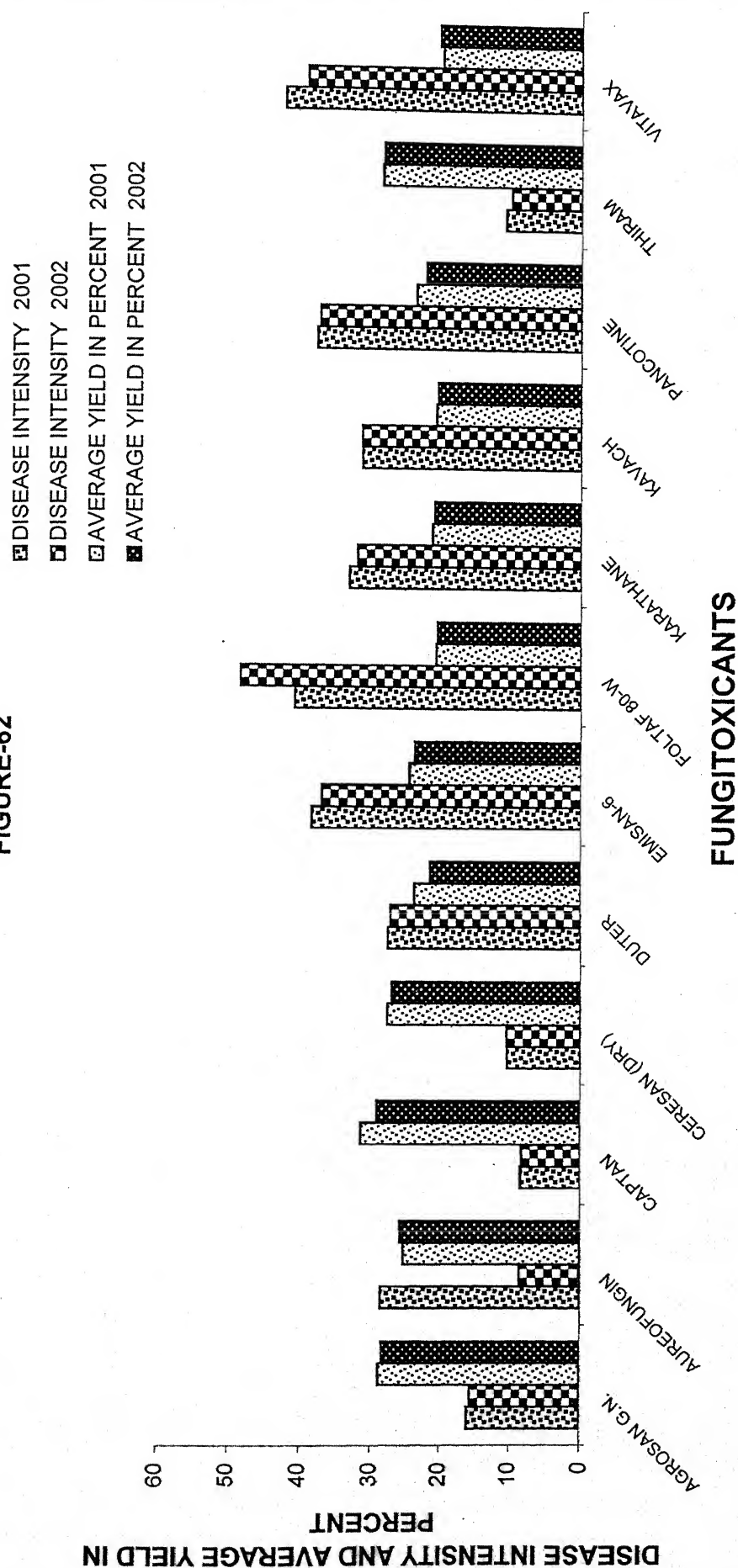
S. No.	Fungi-toxicants	Dose (%)	Disease Intensity (%)		Percent reduction in disease intensity over control		Average yield (%)		Percent yield increase over control	
			2001	2002	2001	2002	2001	2002	2001	2002
1.	Agrosan G.N.	0.2	15.98	15.49	64.20	63.70	28.85	28.35	30.30	36.57
2.	Aureofungin	0.1	28.54	8.57	31.71	38.37	25.27	25.72	15.87	23.74
3.	Captan	0.2	8.42	8.25	80.22	81.49	31.34	29.08	43.59	39.29
4.	Ceresan (Dry)	0.2	10.38	10.44	65.31	65.42	27.60	26.94	26.05	31.34
5.	Duter	0.1	27.59	27.18	34.53	34.68	23.70	21.50	8.07	22.14
6.	Emisan-6	0.2	38.39	36.94	9.65	9.29	24.51	23.70	11.74	12.80
7.	Foltaf 80-W	0.2	40.78	48.35	4.98	5.79	20.70	20.54	4.52	16.57
8.	Karathane	0.2	33.24	32.07	38.64	23.41	21.30	21.00	2.68	29.81
9.	Kavach	0.2	31.43	31.50	37.69	21.42	20.80	20.60	2.16	28.74
10.	Pancotine	0.2	37.86	37.48	10.96	9.73	23.72	22.28	9.59	6.37
11.	Thiram	0.2	10.77	9.97	74.68	78.15	28.63	28.47	33.05	36.47
12.	Vitavax	0.2	42.39	39.24	4.63	6.27	19.94	20.35	5.92	2.39
13.	Control	—	44.55	41.64	—	—	19.62	19.82	—	—

FIGURE -62

Effect of spraying of Fungitoxicants on the intensity of leaf Spot disease caused by *Alternaria alternata* (Fries.), Keissler, of Dolichos bean (*Dolichos lablab*, L.) in Pot Experiments.

1. Agrosan G.N.
2. Aureofungin.
3. Captan.
4. Ceresan (Dry).
5. Duter.
6. Emisan-6.
7. Foltaf 80-W.
8. Karathane.
9. Kavach.
10. Pancotin.
11. Thiram.
12. Vitavax.

FIGURE-62

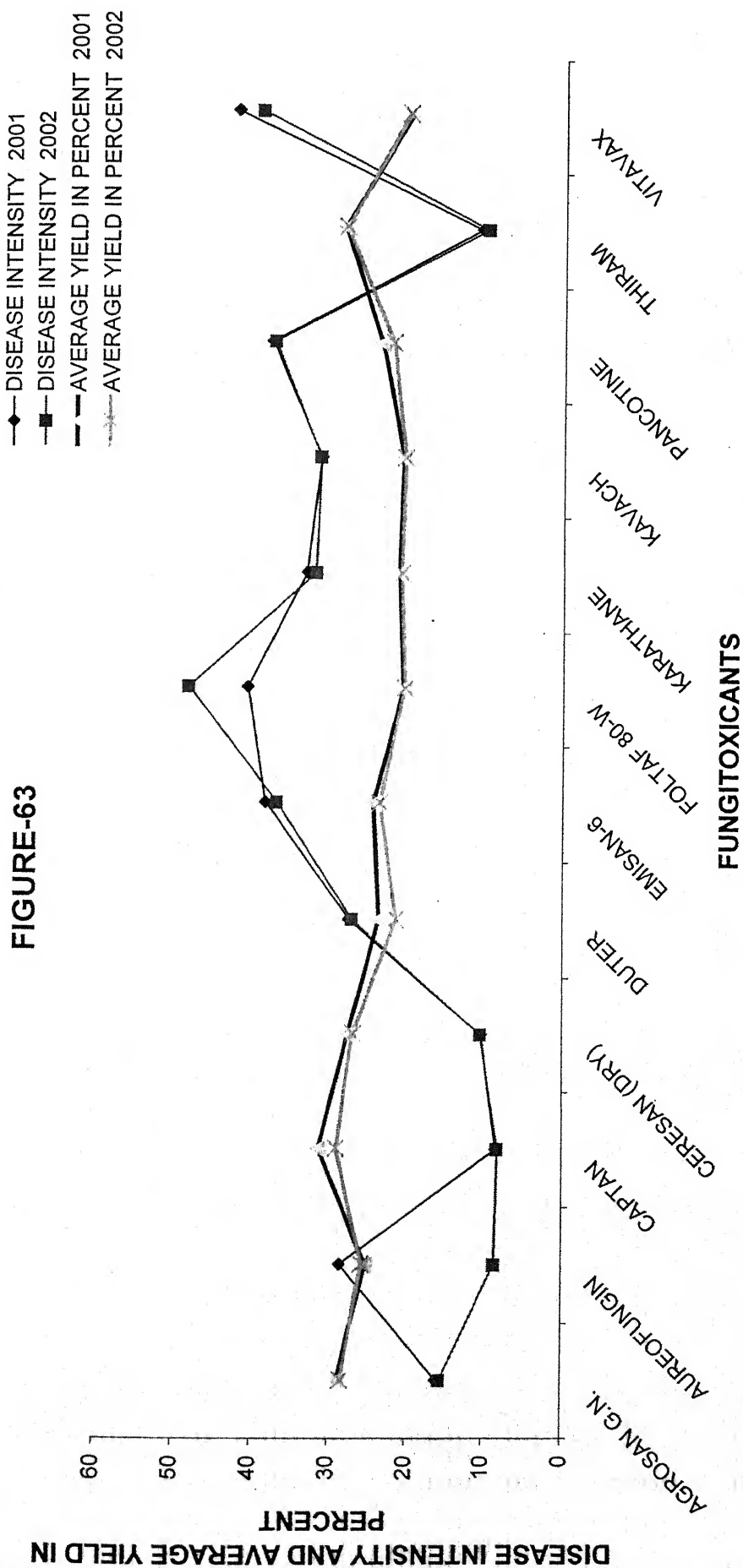


EFFECT OF SPRAYING OF FUNGITOXICANTS ON THE INTENSITY OF LEAF SPOT DISEASE CAUSED BY *Alternaria alternata* (Fries.), Keissler, OF DOLICHOS BEAN (*Dolichos lablab*, L.) IN POT EXPERIMENTS.

FIGURE -63

Effect of spraying of Fungitoxicants on the intensity of leaf Spot disease caused by *Alternaria alternata* (Fries.), Keissler, of Dolichos bean (*Dolichos lablab*, L.) in Pot Experiments.

1. Agrosan G.N.
2. Aureofungin.
3. Captan.
4. Ceresan (Dry).
5. Duter.
6. Emisan-6.
7. Foltaf 80-W.
8. Karathane.
9. Kavach.
10. Pancotin.
11. Thiram.
12. Vitavax.



EFFECT OF SPRAYING OF FUNGITOXICANTS ON THE INTENSITY OF LEAF SPOT DISEASE CAUSED BY *Alternaria alternata* (Fries.), Keissler, OF DOLICHOS BEAN (*Dolichos lablab*, L.) IN POT EXPERIMENTS.

Foltaf-80W. Foltaf 80-W fungitoxicant was found poorest amongst all the fungitoxicants tested in controlling the disease in both the years 2001 and 2002. The highest disease intensity was recorded in control, where no fungitoxicants were used.

Further, it was also recorded that plants treated with Captan exhibited significantly better yield 31.34 gms. and 29.08 gms. per plant than control weighing 19.62 gms. and 19.82 gms. per plant in the years 2001 and 2002. The next 28.63 gms. and 28.47 gms. per plant yield was found in the treatment with Thiram followed by Agrosan G.N., Ceresan (Dry), Aureofungin (Antibiotic), Emisan-6, Pancotine, Duter Karathane and Kavach. However the lowest yield weighing 19.49 gms. and 20.35 gms., per plant was recorded in Vitavax, followed by control, which exhibited 19.62 gms. and 19.82 gms., per plant respectively.

2. EVALUATION OF FUNGICIDES IN THE FIELD

The efficacy of same sets of fungitoxicants as done in pot experiment was further examined under field conditions in order to detect out their effectiveness in reducing the disease severity and improving the yield, during the two consecutive years 2001 and 2002 and results recorded are summarised in Table XXXXX and corresponding Figures 64 and 65.

It is evident from the results obtained that all the fungitoxicants examined were found significantly superior to control. The application of Captan was found to be most effective in controlling the disease up to the extent of 83.68 per cent and 79.29 per cent in both the years 2001 and 2002 and proved significantly superior to other fungitoxicants tested. The next in effectiveness was Thiram, which controlled the disease up to 81.78 per cent and 83.47 per cent respectively in both the years 2001 and 2002 followed by Vitavax, Ceresan (Dry), Agrosan G.N., Aureofungin (Antibiotic), Duter, Karathane, Kavach, Emisan-6 and Pancotine. Agrosan G.N., Aureofungin (Antibiotic), Duter, Emisan-6, Pancotine, Foltaf-80-W, Karathane and Kavach differed significantly in the years 2001 and 2002 respectively. Foltaf-80-W fungitoxicant, was observed poorest amongst all the fungitoxicants under study in controlling the disease in both the years 2001

TABLE - XXXXX

Effect of spraying of Fungitoxicants on the intensity of leaf Spot disease caused by *Alternaria alternata* (Fries.), Keissler, of Dolichos bean (*Dolichos lablab*, L.) Field Experiments.

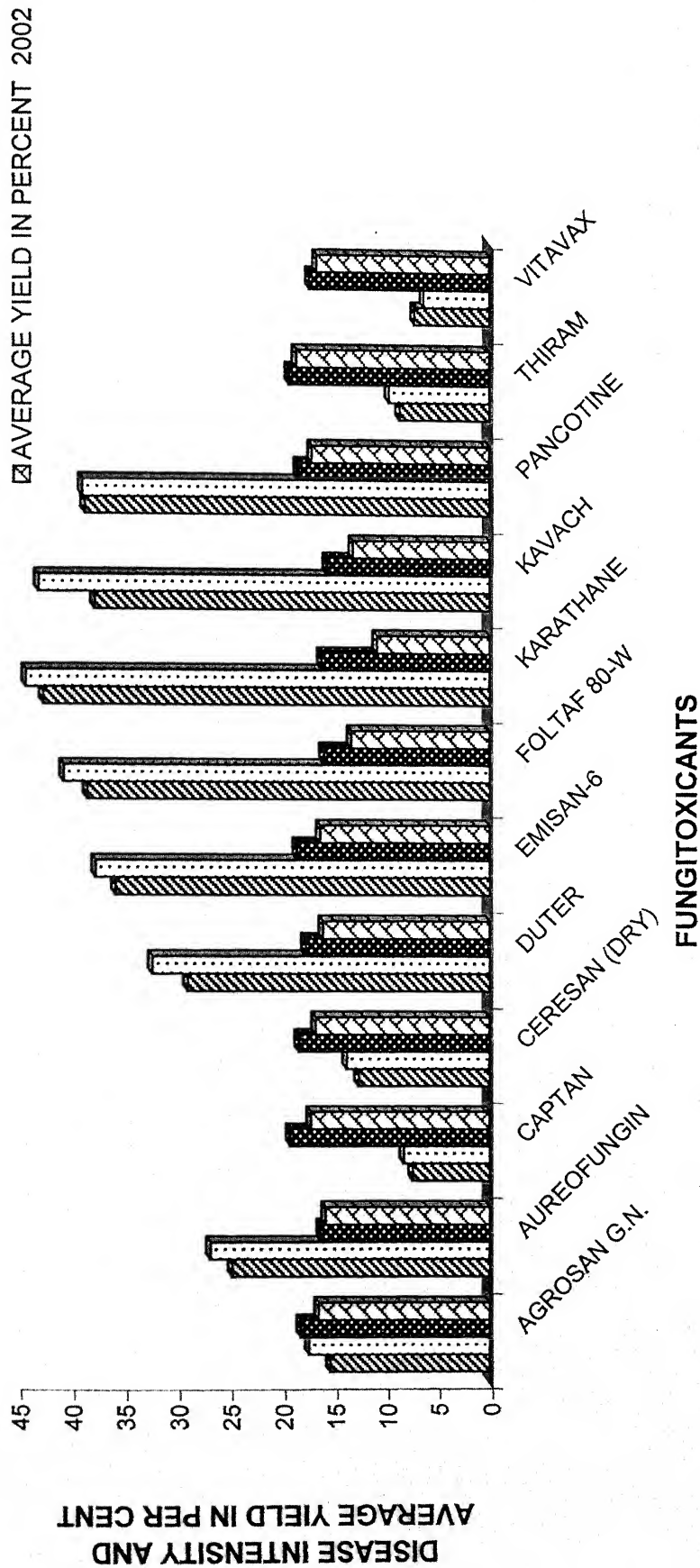
S. No.	Fungi-toxicants	Dose (%)	Disease Intensity (%)		Percent reduction in disease intensity over control		Average yield (%)		Percent yield increase over control	
			2001	2002	2001	2002	2001	2002	2001	2002
1.	Agrosan G.N.	0.2	15.39	17.45	68.39	65.14	18.20	16.54	14.36	26.18
2.	Aureofungin	0.1	24.75	26.84	48.72	46.87	16.34	15.85	6.50	3.94
3.	Captan	0.2	7.43	8.29	83.86	79.29	19.25	17.32	22.25	20.00
4.	Ceresan (Dry)	0.2	12.68	13.85	74.51	72.77	18.44	16.87	15.05	27.20
5.	Duter	0.1	29.09	32.37	39.97	35.97	17.83	16.13	14.35	13.10
6.	Emisan-6	0.2	35.84	37.76	25.58	25.15	18.64	16.40	15.79	20.91
7.	Foltaf 80-W	0.2	38.58	40.80	19.78	18.82	16.08	13.42	4.42	5.73
8.	Karathane	0.2	42.73	44.37	11.67	12.72	16.29	10.93	9.07	8.87
9.	Kavach	0.2	37.92	43.18	11.25	12.47	15.83	13.27	15.09	20.45
10.	Pancotine	0.2	38.84	39.06	19.91	22.55	18.59	17.30	8.59	19.83
11.	Thiram	0.2	8.73	9.75	81.78	83.47	19.47	18.79	21.62	28.84
12.	Vitavax	0.2	7.29	6.39	81.77	80.32	17.58	16.85	19.70	13.34
13.	Control	-	48.14	50.37	89.92	-	15.87	14.75	-	-

FIGURE -64

Effect of spraying of Fungitoxicants on the intensity of leaf Spot disease caused by *Alternaria alternata* (Fries.), Keissler, of Dolichos bean (*Dolichos lablab*, L.) in Field Experiments.

1. Agrosan G.N.
2. Aureofungin.
3. Captan.
4. Ceresan (Dry).
5. Duter.
6. Emisan-6.
7. Foltaf 80-W.
8. Karathane.
9. Kavach.
10. Pancotin.
11. Thiram.
12. Vitavax.

FIGURE-64



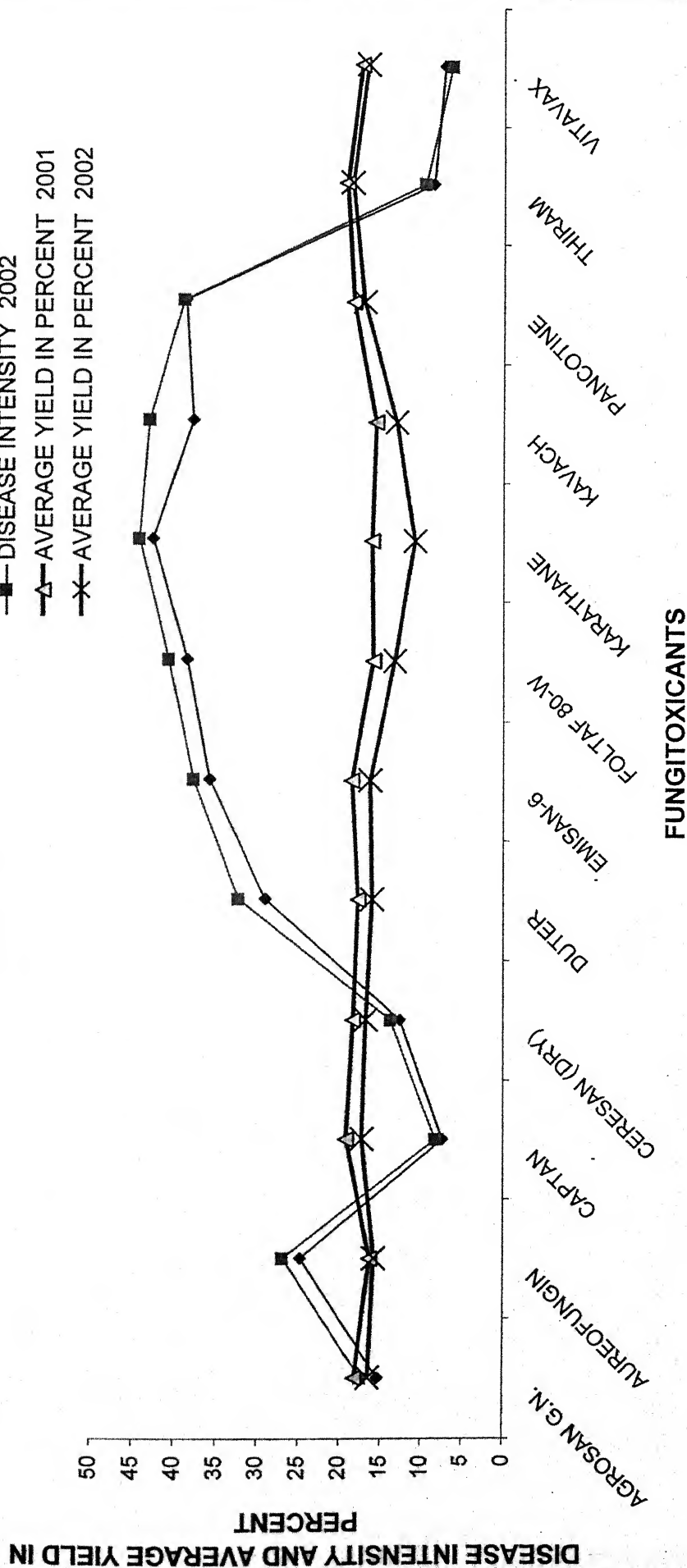
EFFECT OF SPRAYING OF FUNGITOXICANTS ON THE INTENSITY OF LEAF SPOT DISEASE CAUSED BY *Alternaria alternata* (Fries.), Keissler, OF DOLICHOS BEAN (*Dolichos lablab*, L.) IN FIELD EXPERIMENTS.

FIGURE -65

Effect of spraying of Fungitoxicants on the intensity of leaf Spot disease caused by *Alternaria alternata* (Fries.), Keissler, of Dolichos bean (*Dolichos lablab*, L.) in Field Experiments.

1. Agrosan G.N.
2. Aureofungin.
3. Captan.
4. Ceresan (Dry).
5. Duter.
6. Emisan-6.
7. Foltaf 80-W.
8. Karathane.
9. Kavach.
10. Pancotin.
11. Thiram.
12. Vitavax.

FIGURE-65



EFFECT OF SPRAYING OF FUNGITOXICANTS ON THE INTENSITY OF LEAF SPOT DISEASE CAUSED BY *Alternaria alternata* (Fries.), Keissler, OF DOLICHOS BEAN (*Dolichos lablab*, L.) IN FIELD EXPERIMENTS

and 2002. The highest disease intensity 48.14 per cent and 50.37 per cent was recorded in control, where no fungitoxicants were applied in both the years 2001 and 2002.

Further, it was also recorded that plots treated with Captan gave the significant yield of 19.25 gms., and 17.32 gms, per plant in comparison to other treated and untreated plots in both the years 2001 and 2002 respectively. The next fungitoxicant in superiority was Thiram, which yield 19.47 gms., and 18.79 gms., per plant in both years respectively, followed by Ceresan (Dry), Agrosan G.N., Pancotine, Duter, Emisan-6, Vitavax, Karathane, Kavach, Aureofungin (Antibiotic) and Foltaf-80-W. However, the lowest yield of 16.08 gms. and 13.42 gms., per plant was found in treatment with Foltaf-80-W, respectively in both the years. The yield differences intreated and untreated plots in general in the year 2001 was significant in comparison to the year 2002. Emisan-6 was also found effective in increasing the yield in both the years.

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Chapter - V
DISCUSSION

DISCUSSION

Dolichos bean (*Dolichos lablab*, L.) commonly known as "Sem" is one of the important vegetable yielding crop grown in India, like many other crops. It is also subjected to attack by a number of diseases, which are great menace in augmenting its production. It has been observed that with the change in varietal pattern and cultural practices in many cases even unimportant diseases assume epidemic proportions and new diseases make their appearance. It appears to hold good for leaf spot of Dolichos bean (*Dolichos lablab*, L.), caused by *Alternaria alternata* (Fries.), Keissler, which is one of the important diseases of Dolichos bean, has been reported to cause considerable losses to this crop during the recent years, although several efforts have been made during the recent years for its improvement due to its importance in agriculture economy, as well as in vegetable production but the yield of this crop is still quite low as compared to many other countries of the world. If the production of this crop is to be increased, the Scientists of various disciplines will have to work hard and the plant protection measures, will have to be given due importance. In view of the fact very meagre information is available, so far on this disease, detailed studies, were taken up on various aspects of the pathogen and pathogen to fill up the lacuna in our knowledge for planning suitable strategy in order to manage this malady effectively.

The occurrence of the disease during the course of survey at Vegetable Research Farm, Kalyanpur, Kanpur and Chandra Shekhar Azad University of Agriculture and Technology, Kanpur, were found to be quite prevalent during the Kharif crop seasons, during the years 2001 and 2002. With a view to ascertain the status of this disease in various important Dolichos bean growing areas of U.P., the surveys, were conducted in 2001 and 2002, which revealed that leaf spot of Dolichos bean, was widely prevalent.

The disease incidence in different localities from different

germplasms/ cultures viz; Culture-7301, Kamgranj Selection-2, Culture-9012, Culture-7015, Culture-9118, Culture-7708, HD-4, Akra Jai, DB-1, Culture-8403, Todi-125-136, Culture-7604, Pusa Early Prolific, Goyal; JDL-79, Rajani, Culture-6801, Culture-8101, Culture-7710, Hatikan, HD-66, Culture-7210, Culture-9109, Culture-9113, Culture-8005, TDL-85, HD-93 and Kalyanpur Type - 1 varied from 13.48 per cent to 31.57 per cent and 10.25 per cent to 34.85 per cent from the germplasms/cultures viz., Culture-7301, Culture-7210, Akra Jai, Culture-9118, Culture-6801, Haikan, Culture-8005, Culture-7015, Todi-125-126, Goya, JDL-85, Pusa Early Prolific, Culture-9102, Kamgranj Selection-2, HD-4, Culture-9109, Culture-8101, HD-66, DPL-1, Culture-8403, DB-1, HD-93, Rajani, JDL-79, Culture-7604, Culture-7710, Culture-9113, Culture-9104 and Kalyanpur Type - 1 in Kharif season of the year 2001 and 2002 respectively showing its wide spread occurrence. The maximum disease incidence 31.57 per cent and 34.85 per cent was recorded at Vegetable Research Farm Chandra Shekhar Azad University of Agriculture and Technology, Kanpur in both the years from the germplasm/culture, "Kalyanpur Type - 1", followed by 30.80 per cent and 29.26 per cent at Government Agriculture Centre, Jalaun and Crop Research Farm Modipuram Area, Meerut from the germplasms/cultures Culture HD-93 and Culture-9113 and rest of locations, while minimum 13.48 per cent and 10.25 per cent disease incidence, was recorded at Agriculture Science Centre, Ganiwa, Chitrakoot in both the years.

The average disease incidence ranged from 13.48 per cent to 31.57 per cent and 10.25 per cent to 34.85 per cent in both the years of survey at different Crop Research Stations. In general, it was concluded that minimum disease incidence was recorded at Agriculture Science Centre, Ganiwa, Chitrakoot.

The intensity of disease varied from 22.40 per cent to 38.40 per cent from the germplasms/cultures viz; Todi-125-126, JDL-79, Culture-9113, Culture-7210, Akra Jai, Culture-6801, Hatikan; Culture-9102, Culture-7301,

Culture-7708, Goya, Culture-8005, Culture-9118, HD-66, Culture-7710, Culture-8403, DB-1, HD-4, Pusa Early Prolific, Kamgranj Selection-2; HD-93, Culture-7015, Culture-8101, Rajani, JDL-85, Culture-9109, Culture-7604 and Kalyanpur Type - 1 and 17.90 - 40.50 per cent from the germplasms/cultures viz., Culture-7710, Culture-9118, Culture-9102, Hatikan, Culture-9104, Akra Jai, Culture-7301, Culture-7604, Culture-9109, DPL-1, Culture-8005, Kamgranj-Selection-2, Goya, Culture-6801, HD-93, Pusa Early Prolific, JDL-79, Rajani, Culture-8403, Culture-7015, DB-1, HD-4, JDL-85, HD-66, Culture-9113, Todi 125-136, Culture-7210, Culture-8101 and Kalyanpur Type-1 in the years 2001 and 2002. Maximum 38.40 per cent and 40.50 per cent disease intensity was recorded from the germplasm/culture "Kalyanpur Type-1" at Chandra Shekhar Azad University of Agriculture and Technology, Kanpur, followed by 37.80 per cent and 39.50 per cent from Oil Seed Research Farm, Kalyanpur Kanpur and Crop Research Farm Saraimira, Farrukhabad from the germplasm/cultures 7604 and 8101 and rest of locations in both the years, 2001 and 2002, while minimum 22.40 per cent and 17.90 per cent disease intensity was recorded from Directorate of Vegetable Research Farm, Varanasi and Government Agriculture Centre, Atarra, Banda from the germplasms/cultures Todi-125-126 and Culture-7710 in both the years respectively.

In general the maximum disease intensity was recorded from Chandra Shekhar Azad University of Agriculture and Technology, Kanpur.

The disease incidence as well as disease intensity could not be recorded in the years 2001 and 2002 at Crop Research Farm, Deegh, Kanpur Dehat and Regional Agriculture Research Station Dilip Nagar, Kanpur and Regional Research Centre, Amroha, Jhansi, respectively.

Alternaria tenuis, was for the first time reported from India by Bose (1942) on the leaves of Sunflower and it was considered to be a severe and destructive pathogen. Goyal (1966), from Rajasthan also reported this disease caused by *Alternaria alternata* to be quite prevalent in Rajasthan.

Estimation of intensity percentage of wheat leaf blight caused by *Alternaria triticina*, was made by Verma *et al.* (1969). Narain and Saksena (1981), also reported that *Alternaria alternata*, affects all the aerial plants parts including leaves, head and seed. Effect of seasonal variations on the severity of *Alternaria* leaf spot of Sunflower, was studied by Raut *et al.* (1985) in Vidharbha region of Maharashtra and they found 33, 17 and 6.0 per cent disease intensity in Kharif season. The results of these workers support the present findings. However the Head and Stem infection of Sunflower plants as observed by Narain and Saksena (1981), were not noticed during the present investigation, which may be due to existence of varied virulence of the pathogen in nature. Shtienberg (1992), studied the variable associated with intensity of *Alternaria* leaf spot of Cotton.

Studies on the disease symptology revealed that, it appeared as numerous small spots on the upper surface of leaves. The spots were recorded as brown to black, circular to oval with paler margins and yellow halo, measuring 0.15 to 1.0 cm. in size with characteristic concentric rings and cracked centre. The spots at first were, recorded as smaller in size. In case of severe attack, the spots were found numerous extending alongwith the whole leaf surface due to coalescence of adjacent spots. In late stage of the disease, the affected spots became perforated irregularly due to falling away of dead tissue causing "Short holes". These symptoms, were found similar to those as described by Bose (1942) and Goyal (1966). More or less similar symptoms were also reported by Gupta (1985), on Broad bean (*Vicia faba*, L.).

In pathogenicity test the fungus produced symptoms, similar to those observed in nature on both injured and un-injured leaves of Dolichos bean. The disease symptoms, were developed within ten days after inoculation with comparatively higher disease intensity in the former case, which indicated that the pathogen was a potential one and that injury favoured disease development. The fungus, was reisolated from the infected

leaves and was found identical with the original isolates. The pathogenicity of the fungus was established by other workers also on several including Sunflower (Bose, 1942), Linseed (Siddiqui, 1963), Cotton (Rao, 1965), Tomato (Tandon and Chaturvedi, 1965), Dolichos bean (Goyal, 1966), Barley (Dhanraj, 1970), Tobacco (Staveland and Slana, 1971), Rape (Vaartnou and Tewari, 1972) and Narain and Saksena (1974) and Arhar (Mehta and Sinha, 1982). Effect of different Crop seasons on the incidence of *Alternaria helianthi* in Sunflower seeds, was studied by Kaur *et al.* (1991). Their findings support the pathogenic nature of the fungus as observed in the present work.

The morphological characters of the fungus, studied on Potato Dextrose agar medium revealed that the colonies were moderately fast growing in the beginning dull-white; fluffy; circular and later turned into dark and greenish olive with abundant sporulation. Mycelium was septate; branched in earlier stages; hyaline; later black olive buff and measured 3.20 - 8.60 μ in width. Conidiophores were found developing singly or in groups; simple; septate; straight or bent; sometimes branched; swollen terminally; geniculate; dark olive buff and measuring 24.50 - 69.30 \times 3.20 - 6.40 μ in size. Conidia, were observed in chains of 3-21 as muriform, ovoid to obclavate, or obpyriform; dark olive buff in colour; smooth; sometimes verrucose; formed in chains; with 1-6 transverse septa and 0-6 longitudinal septa measuring 16.50 - 40.90 \times 8.20 - 12.50 μ . Beaks were short, light olive buff in colour; conical or cylindrical and measuring 4.60 - 16.90 \times 3.40 - 5.80 μ in size with 0-2 transverse septa. Chlamydospores were terminal; intercalary and dark olive buff in colour measuring 12.70 - 22.80 μ in diameter. The morphological characters of the pathogen observed in the present study, are more or less similar as described by Mason (1928), Keissler (1912), Dey (1933), Bose (1942), Groves and Skolko (1944), Neergaard (1945), Kamal (1950), Swank (1951), Arya and Prasad (1952), Kapoor and Hingorani (1958), Malone and Muskett (1964), Simmons (1967) and Ellis (1971) for various isolates of *Alternaria alternata* and it was

identified as such.

The effect of different solid media on the growth of the pathogen revealed that Potato dextrose agar, Richard's agar and Czapek's agar media supported best growth of pathogen. Good growth was recorded on Coon's agar; Leaf decoction agar; fair growth on Corn meal agar; Malt salt agar and Sabouraud's agar media and least minimum growth on Brown's agar medium. Excellent sporulation on Potato dextrose agar, Czapek's agar and Richard's agar media; good sporulation on Coon's agar and leaf decoction agar media and fair on Corn meal agar, Malt salt agar and Sabouraud's agar media. Poor sporulation was observed on Asthana and Hawker's agar medium and no sporulation was observed on Brown's agar medium. These observations are in conformity with the findings of Lilly and Barnett (1951), who suggested the superiority of natural media over synthetic media in relation to mycelial growth of fungus due to the fact that the former contains more nutrient than the later. Arya and Prasad (1952), found maximum growth and sporulation of *Alternaria brassicae*, and *A. macrospora* (*A. alternata*) on Linseed host leaf extract. Hawker (1956), also observed that mycelial growth was favoured by nutritional factors. Gupta and Uprety (1964), found better growth of *Alternaria alternata* and sporulation on decoction containing Tamarind (*Tamarindus indica*), *Cassia fistula*, *C. obtusifolia* and *Mangifera indica* on Potato dextrose agar medium. Ashour and El_Kadi (1958), Goyal (1977), Allen *et al.* (1983) and Xu *et al.* (1984), reported that the Potato dextrose agar medium followed by Richards agar medium supported maximum growth and sporulation of *A. alternata*. Similar results were also observed by Rangaswamy and Sambandan (1960), Xu *et al.* (1984) and Chattannavar *et al.* (1987). By Ionnaidis and Main (1973), Lima Bean agar medium and by Chandrashekhar and Ball (1980), Malt extract agar medium, were recorded as best medium for growth and sporulation of *A. alternata*. Mathur and Sarbhoy (1977), also observed that out of six different Synthetic media tried, Richard's agar medium supported maximum growth of pathogen but contrary to the present findings, they reported poor sporulation on this medium. They

also found least growth and sporulation of *A. alternata* on Asthana and Hawker's medium, which is in accordance with the present findings. Susuri and Hagedorn (1986), also reported that *A. alternata* grew well on Potato dextrose agar medium.

In studies of the cultural characters of the pathogen on ten solid media used, it was found that the pathogen showed marked variation in growth and colony characters on different media. The growth was good; compact; dark and greenish with circular colony on Potato dextrose agar medium. More or less similar characters, were observed on Richard's agar medium. The colony growth was circular on all the media except Czapek's agar medium on which lobed growth was observed. On the rest of the media, it was dark; greenish; light green; creamy white; smoke black with fair yellowish growth and zonation, which is sometimes distinct or not having clear zonation. The colonies were found circular; compact and olivaceous black having fair growth and distinct zonation on Malt extract agar medium. Pigmentation was found absent in all the media tested.

Colour of hyphae was recorded as olive buff on Czapek's agar; Richard's agar and Sabouraud's agar media; mid olive on Brown's agar media; olive buff to greyish on Malt salt agar medium; dark olive buff on Asthana and Hawker's agar medium; pale olive grey on Coon's agar medium; light olive grey on Corn meal agar and Leaf decoction agar media and colourless to greyish on Potato dextrose agar medium. Hyphae were found septate and varied in size from 2.50 - 8.0 μ in width on different types of media.

Conidia were also found variable in colour viz; olivaceous to dark brown on Czapek's agar, Malt salt agar, Richard's agar and Sabouraud's agar media; olivaceous to brown on Potato dextrose agar medium; dark olive brown on Asthana and Hawker's agar medium; olive buff to brown on Brown's agar medium; light brown on Coon's agar medium; dark olive grey on Corn meal agar medium and deep olive brown on Leaf decoction agar

medium.

Colour of conidiophores, was recorded as olive buff to brown on Brown's agar and Richard's agar media; mid olive to brown on Czapek's agar and Sabouraud's agar media; olivaceous brown on Coon's agar and Malt salt agar media; dark olive buff on Asthana and Hawker's agar medium; pale to olive brown on Potato dextrose agar medium and olive grey on Corn meal agar medium.

The conidiophores were recorded varying in size from $24.20 - 68.60 \times 3.10 - 7.80 \mu$ on different types of media. Number of conidia borne on conidiophores in chains was also found variable on different types of media viz; 2-4 (Asthana and Hawker's agar, Brown's agar and Coon's agar media); 2-5 (Corn meal agar, Czapek's agar and Sabouraud's agar media); 2-6 (Leaf decoction agar and Malt salt agar media); 2-7 (Richard's agar medium) and 3-7 (Potato dextrose agar medium). Septation in conidia, was also observed variable on different types of media. Transverse septa varied from 1-5 to 2-8 viz; 1-4 (Corn meal agar media); 1-5 (Asthana and Hawker's agar and Brown's agar media); 2-7 (Coon's agar, Czapek's agar, Leaf decoction agar, Malt salt agar and Sabouraud's agar media) and 2-8 (Potato dextrose agar and Richard's agar media), while longitudinal septa varied from 0-3 to 0-6 viz; 0-3 (Asthana and Hawker's, Brown's agar, Coon's agar and Corn meal agar media); 0-4 on (Czapek's agar, Leaf decoction agar. Malt salt agar and Sabouraud's agar media); 0-5 on Richard's agar medium and 0-6 on Potato dextrose agar medium. Conidia were also found varying in size from $7.0 - 35.0 \times 3.0 \times 13.80 \mu$ on different types of media.

The beaks were found septate and varied in size from $10.0 - 78.40 \times 1.20 - 6.40 \mu$ on different types of media. The transverse septa varied from 0-2 viz; 0-1 (Asthana and Hawker's agar, Czapek's agar, Leaf decoction agar, Malt salt agar and Sabouraud's agar media) and 0-2 (Brown's agar, Coon's agar, Corn Meal agar, Potato dextrose agar and Richard's agar media). The beaks were also variable in colour viz; Olive green (Asthana and Hawker's

agar medium); olive buff to brown (Czapek's agar medium); olive grey (Corn meal agar medium); dark brown (Czapek's agar and Sabouraud's agar media); dark olive brown (Leaf decoction agar medium); olive to dark brown (Malt salt agar medium) and light olive buff (Potato dextrose agar medium). The beaks were also found varying in shape viz; cylindrical on Corn meal agar, Czapek's agar leaf decoction agar, Richard's agar and Sabouraud's agar media and conical on Asthana and Hawker's agar, Brown's agar, Coon's agar, Malt salt agar and Potato dextrose agar media.

These findings agree with the observations of Neergaard (1945) and Gupta *et al.* (1969), who had studied the effect of culture medium on morphological characters of the pathogen and recorded cultural characters and spore morphology of *Alternaria*, influenced by the constituents of substrate media. Swank (1951) and Kapoor and Hingorani (1958), reported good and dark grey growth of *Alternaria alternata* on Potato dextrose agar medium. Dhanraj (1970), also observed good growth of *A. alternata* on Potato dextrose agar medium. Wide variations in morphological and cultural characters of *A. alternata*, isolated from different hosts were also studied by Dhanraj (1970), Reddy and Bilgrami (1974), Ram and Gupta (1975), Madaun *et al.* (1979) and Melkania (1980). Sokhi *et al.* (1972), studied cultural and morphological characters of *Alternaria alternata* isolated from Wheat and found bigger spores on host as compared to Malt salt agar medium. Siddiqui (1963) and Mathur and Sarbhoy (1977), observed a marked variation in vegetative parts of *A. alternata* on different media used. Shukla (1981), recorded more or less similar cultural observations for the isolate of *A. alternata* on Potato dextrose agar, Corn meal agar, Malt extract agar Czapek's (Dox) agar, Kirchoof's agar, Richard's and Sabouraud's agar media. These findings provide further credence to the results of Neergaard (1945).

Chlamydospores were found terminal as well as intercalary varying in size 4.70 - 22.80 μ in diameter on all the different types of media under study but varying in colour viz; dark brown on Czapek's agar and Sabouraud's agar media; dark olive green on Asthana and Hawker's agar

medium; dark olive brown on Leaf decoction agar medium; olivaceous to dark brown on Malt salt agar medium; olive buff on Potato dextrose agar medium; light brown on Coon's agar and Richard's agar media and olive grey on Corn meal agar medium. The present findings are in agreement with the results of Narain *et al.* (1991) and Lagopodi and Thanassouloupoulos (1995).

Out of the ten liquid media tested the best growth of fungus was obtained on Potato dextrose medium, followed by Richard's and Czapek's media; good on Coon's and Sabouraud's media; fair on Asthana and Hawker's, Leaf decoction and Malt salt media, whereas Brown's medium supported poor growth. Sporulation of the pathogen as excellent was observed on Potato dextrose; Richard's and Czapek's media; while good sporulation on Coon's and Sabouraud's media, fair on Asthana and Hawker's, Leaf decoction and Malt salt media; and poor sporulation was recorded on Brown's medium. It was remarkable to note that more or less similar results were recorded in various media included in the present study in both solid and liquid states by Ashour and El-Kadi (1959), Rangaswami and Sambandan (1960), Goyal (1977), Mathur and Sarbhoy (1977) and Xu *et al.* (1984).

The studies taken up to find out the optimum temperature for the growth and sporulation of the pathogen, it was observed that *Alternaria alternata*, could grow over a temperature range of 5°C (T1) to 50°C (T-10) and the optimum temperature for its growth was found to be 30°C (T-6), whereas the minimum at which it could grow was 5°C (T-1). Excellent sporulation of the pathogen was noticed at 30°C (T-6); 35°C (T-7) and 25°C (T-5); good at 40°C (T-8) and 20°C (T-4); fair at 15°C (T-3); poor at 10°C (T-2) and 45°C (T-9), whereas no sporulation was observed at 5°C (T-1) and 50°C (T-10). The optimum and the maximum temperatures for the growth of *A. alternata* were 35°C (T-7) and 25°C (T-5) and 30°C (T-6) respectively. The present results are in accordance with the results of earlier workers like Arya and Prasad (1952), Ashour and El-Kadi (1958), Tandon (1961), Siddiqui (1963), Verma (1963), Csyzewska (1970), Crissan and Meseson (1970), Kamal *et al.* (1971), Griyorju (1976) and Mathur and Sarbhoy (1977), who have

worked on *A. alternata* isolated either from Cotton or other hosts. Xu *et al.* (1984) and Chattannavar *et al.* (1987), obtained maximum growth of the fungus on pH 6.50 and temperature 20°C, which is very close to the present findings, while Saad and Hagedorn (1970), observed little growth of the fungus at 4°C. Vakalounakis and Malathrakis (1988), found the mycelial growth of *A. alternata* at 5°C - 40°C, with optimum temperature at 26°C. Except for slight variation, which may be explained due to certain differences. The results of all these workers supported the present findings.

Studies on the pH requirement of the pathogen revealed that it could grow over a wide range of pH values from 2.50 (P-1) to 12.0 (P-20) and its optimum pH for its growth was found to be 6.50 (P-9). The poorest growth of the pathogen was recorded at pH-12 (P- 20). The growth of the pathogen increased from pH 3.0 (P-2) to pH 6.50 (P-9), beyond which the growth started decreasing. Excellent sporulation of the fungus was observed at the pH levels of 6.50 (P-9) and 7.0 (P-10); good at pH 7.50 (P-11) and pH 8.0 (P-12); fair at pH 4.50 (P-5); pH 5.0 (P-6), pH 5.50 (P-7), pH 6.0 (P-8), pH 8.50 (P-13), pH 9.0 (P-14) and pH 9.50 (P-15); poor at pH 3.50 (P-3), pH 4.0 (P-4), pH 10.0 (P-16) and pH 10.50 (P-17), whereas no sporulation was observed at pH 2.50 (P-1), pH 3.0 (P-2), pH 11.0 (P-18), pH 11.50 (P-19) and, pH-12.0 (P-20). The present findings are in aggrement with the results of Arya and Prasad (1952), Verma (1969), Xu *et al.* (1984) and Chattannavar *et al.* (1987), who found maximum growth of *A. alternata* at pH 6.50. Saad and Hagedorn (1970), reported the optimum pH requirements for the growth and sporulation of the two isolates of *A. alternata* to be 6.60 and 6.50 respectively. In addition the observations of other workers (Ashour and El-Kadi, 1959; Goyal, 1977; Mathur and Sarbhoy, 1977; Singh *et al.*, 1980 and Mehta and Brogin 2000), *A. alternata* is capable of growing over a wide pH range, also corroborate the results of present study. The present findings further indicated that culture of the fungus, changed the initial value of the medium, which may be attributed to the metabolic activity of the growing fungus (Lilly and Barnett, 1951).

Production and activity of enzymes by the pathogen, revealed that *A. alternata*, was capable of producing appreciable amount of Cellulolytic and Pectinolytic enzymes, when grown on Richard's liquid medium supplemented with Carboxymethylcellulose, Sodium polypectate and Citrus pectin as compared to the medium without them, which was clearly indicated by the loss of viscosity of the reaction mixture.

The amount of Cellulase production, was comparatively more than Polygalacturonase enzymes. The present findings are in agreement with the observations of Ruschmann and Bartram (1940), Marsh *et al.* (1947), Tandon and Srivastava (1950), Coulson and Mars (1952), Fischer (1953), Wood (1959), Bateman and Miller (1960), Bateman (1964), Pandey (1965), Hamcock and Mitter (1965, a and b), Prasad (1967), Verma (1969), Egorov *et al.* (1971), Heath and wood (1971), Hasiza (1974), Marimuthu *et al.* (1974), Mehta *et al.* (1975), Tak *et al.* (1985), Miles and Wilcoxson (1984) and Tripathi (1992), who had also reported that *Alternaria alternata* and other species of *Alternaria*, produced these enzymes *in vitro* in Synthetic media. Further Laxminarayana and Reddy (1978), recorded that Carboxymethylcellulose supplemented media were not found conductive for enzymes production and activity in case of *A. alternata*, provide further credence to the present findings. Similarly the production of Cellulase (Cx), Polygalacturonase (PG) and Polymethylgalacturonase (PMG) in presence or absence of substrate by *Alternaria alternata* in the culture, has also been demonstrated by Mehta *et al.* (1974 and 1975), Philip (1976), Mehta (1976), Mehta and Mehta (1979), Kunte and Shastri (1980), Suri and Mandhar (1982), Tak *et al.* (1985) and Singh *et al.* (1991). The presence of these enzymes *in vivo* and their role in disintegration of tissue and cell walls degradation during pathogenesis in the present results are in aggrement with the findings of Marsh *et al.* (1947), Coulson and Mars (1952), Kanovskaya (1966), Patil and Dimond (1967), Reddy and Mahadewan (1967) and Laxminarayana and Reddy (1978), who had reported earlier that *A. tenuis* (*A. alternata*), may decompose cellulolytic

and pectic substances of leaves by attacking both pectin and cellulose. A detailed study on cell wall degrading enzymes was studied by Singh *et al.* (1991).

The plant pathogens of some of the diseases are known to infect at certain age of the host plant, while others may cause infection at any stage of growth. The study on susceptible growth period of Dolichos bean plants to *Alternaria alternata*, it was found that the pathogen could infect the plants of all the ages ranging from 10-90 days, but the maximum disease intensity was recorded in the plants having attained the age of 50 days followed by 60, 40 and 70 days. Disease severity was more in case of older plants than younger ones. It also indicated that pathogen was able to cause infection at any stage of growth and susceptibility of the plants gradually declined with increase in age. The present investigations are in consonance with the observations of Kamal *et al.* (1971), who reported the age of leaves of Cotton per infection to *A. alternata* as 3-15 days and also stated that juice of seedlings and leaves of resistant cultivars inhibited spore germination. Sarkar and Sen Gupta (1978), reported that 20 days old or less old plants were resistant to *A. brassicicola* of Mustard but 30 days old plants, were slightly infected and susceptibility increased with the age of plant. Saad and Hagedorn (1969), Crisan *et al.* (1979), Allen *et al.* (1983), Godnoy and Fernandes (1985) and Singh and Shukla (1986), found that the older leaves and older plants of Sunflower, were more or less susceptible to *A. helianthi*. Singh (1987), recorded the effect of host age and pathogen in productivity of disease. Bedi and Dhiman (1980), recorded maximum incidence of disease in 120 days aged plants and minimum in 20 days aged plants. Suhag *et al.* (1985), found 35 days aged leaves of Radish were more susceptible to *A. alternata* and responsible for multiplication of disease. Chahal (1986), also found that susceptibility of Brown Sarson to *A. brassicae*, which increased with the increase in age of plants. Gupta and Pathak (1986), also observed that Nasik red Onions, were more susceptible to *A. porri*, when inoculated at the age of 60 days. Shukla (1987), also reported that 60 days old plants of

Triticale, were most prone to infection of *A. alternata* as compared to 45 and 30 days old plants. Contrary to these Ghemawat *et al.* (1989), reported that, artificial inoculation of even 5 days old plants of susceptible Sunflower EC-68414 with *A. helianthi* resulted in 100.0 per cent mortality, Sahu *et al.* (1991), recorded the susceptibility of Sunflower plants to *Alternaria helianthi* being influenced by plant age.

The studies on the role of environmental factors (atmospheric temperature, relative humidity and rain fall), revealed that maximum disease, development occurred in the years 2001 and 2002 in the 2nd week of August, when the average temperature was 29.05°C and 28.70°C relative humidity 86.70 and 84.25 per cent and rain fall 0.29 mm. and 11.48 mm. respectively. Thereafter there was gradual decline in disease severity during the month of May and finally by the month of October, it was reduced to traces. The minimum disease intensity was observed in fourth week of October, followed by third week of October, when the atmospheric temperature and relative humidity were found unfavourable.

Further the individual as well as combined effect of relative humidity temperature and rain fall on disease development indicates that most congenial temperatures for maximum disease development were 29.05°C and 28.70°C respectively in the years 2001 and 2002. Similarly the most conducive relative humidity and rain fall were 86.70 per cent and 84.25 per cent and 0.29 mm. and 11.48 mm. in the years 2001 and 2002 respectively. The combined effect indicated that as temperature beyond the congenial ones increased, the disease severity decreased. In case of relative humidity it increased in both the years. These findings are in close proximity with those reported by Dey (1933 and 1948), Arya and Prasad (1952), Mc Fariane *et al.* (1954), Loof (1959), Louvet (1958) and Louvet and Billotte (1964). Arya and Prasad (1952), found that cent per cent development of linseed blight occurred at the temperature range of 26°C - 30°C in the saturated atmosphere for at least 72 hours after inoculation Domach (1957), noticed

that relative humidity of 95.0 per cent to 100.0 per cent and temperature between 21°C and 27°C, was essential for three successive days for development of disease. Ashour and El-Kadi (1958), Kamal *et al.* (1971), Mehta *et al.* (1975) and Ram and Joshi (1978), also reported that atmospheric temperature around 28°C and saturated atmosphere (relative humidity 75.0 per cent to 100.0 per cent), were favourable for disease development. Incidence of disease related to rainfall was studied by Bakos *et al.* (1967). Saad and Hagedorn (1969), studied development of disease at more than 95.0 per cent relative humidity and severe at 16°C, while minimum at 28°C. Successful infection in Virginia Tobacco, was studied by Pamubov (1972) at 20°C and 21°C temperature; high rain fall and more than 67.0 per cent relative humidity.

The effect of ecological factors on sporulation of *Alternaria helianthi* and infection of Sunflower plants was studied by Acimovic (1974). Prasad and Roy (1979), Suhag *et al.* (1985) and Hiremath *et al.* (1988), also observed that infection was intensive between 25°C - 30°C and relative humidity around 80.0 per cent. Allen *et al.* (1983), also found that germination of conidia was favoured at 25°C - 30°C and two hours period of leaf witness, were required to give maximum infection of *A. helianthi* in Sunflower leaves. The most favourable, weather factors in development of leaf spot disease of Brinjal (*A. alternata*), were the temperature and relative humidity ranging between 24°C to 40°C and 47.30 per cent to 51.20 per cent respectively. The leaf spot disease caused by *A. alternata*, was favoured by relative humidity, frequent irrigation and higher application of nitrogen Chattannavar *et al.* (1984). Dingar and Singh (1985) and Ghewande (1986), correlated the temperature between 25°C - 29°C and relative humidity of 87.0 per cent, as favourable for the development of *Alternaria* Leaf spot of Groundnut. Bhargava and Khare (1988), also reported that *Alternaria* blight of Chickpea caused by *A. alternata*, was first seen at flowering stage, when temperature ranged between 25°C - 27°C with relative humidity 80.0 per cent. Hiremath *et al.* (1990), studied correlation between disease severity,

relative humidity and temperature in infection of *A. helianthi* causing Alternaria blight of Sunflower. Infection of *Alternaria alternata*, was favoured moderately at high temperature, normal humidity and dry days (Kumar, 1992). A temperature of 25.90°C - 33.7°C and relative humidity of 89.0 - 95.0 per cent favoured development of Alternaria leaf spot disease of Sunflower (Borker and Patil, 1995).

Meteorological reports showed greater fluctuations during the crop season and was observed that maximum and minimum temperatures and relative humidity varied greatly and affected dew formation. Wilting of the leaf surface, was found to be an important factor for successful infection. Generally leaves start to get wet after sunset and it continued till 8.0 A.M., the next morning. Thus there was a total period of about 16-18 hours of leaf witness. Ram and Joshi (1978), made similar observations of *A. tritici* and stated that successful infection required at least 48 hours in a saturated atmosphere.

Investigation on the most suitable carbon sources for the growth and sporulation of pathogen, *Alternaria alternata*, revealed that sucrose supported its best growth. Good growth was obtained on Galactose, Maltose, Raffinose, Dextrose, Mannose, Fructose and Xylose; fair on Sorbitol, lactose and Mannitol, whereas poor growth was recorded on Dextrin and Rhamnose. Excellent sporulation, was observed on Sucrose, Maltose and Raffinose; good on Sorbitol, lactose and Dextrin, where as poor sporulation resulted on rhamnose. Tandon and Chaturvedi (1963), observed the best growth and sporulation of *A. alternata* on Glucose, followed by Surcose and Maltose. Goyal (1977) reported best growth of the fungus on Maltose, followed Sucrose. Mathur and Sarbhoy (1977) and Thind (1977), recognized the supermacy of Sucrose as the best source of Carbon for growth and sporlation of *A. alternata*. The findings of all these workers confirm the present findings.

In studies to find out the best source of nitrogen for the growth

and sporulation of *A. alternata*, it was observed that amongst twelve organic and inorganic nitrogenous compounds, tested peptone supported the best growth of the pathogen, good growth, was recorded on ammonium chloride, ammonium nitrate, calcium nitrate, potassium nitrate and sodium nitrate. Fair growth was obtained on ammonium acetate, ammonium oxalate and ammonium. Poor growth was exhibited on ammonium carbonate, thiourea and urea. Excellent sporulation was noticed on peptone and sodium nitrate; good on ammonium nitrate, calcium nitrate and potassium nitrate; fair on ammonium acetate, ammonium chloride and ammonium oxalate; poor on ammonium carbonate, ammonium sulphate and urea, whereas no sporulation was recorded in case of thiourea and control. These observations are in complete agreement with the findings of Tandon and Chaturvedi (1963), who observed that nitrates of Potassium, Sodium and Calcium, were good sources for the growth of *A. alternata*, while urea supported poor growth. According to Shukla (1981), Susuri and Hagedorn (1986), peptone was a good source of nitrogen for the growth of *A. alternata*, which provide further credence to the present findings.

The effect of different doses of nitrogen, phosphorus and potash on the severity of leaf spot disease of Dolichos bean, revealed that disease severity and plant susceptibility increased with the increase in the level of nitrogen, whereas negative correlation, was observed with the level of phosphorus and potash with severity of the disease.

In respect of the effect of nitrogen, phosphorus and potash on disease severity, it was observed that 60 Kg; P_2O_5 + 40 Kg. K_2O , were more effective in reducing the disease intensity as compared to 60 Kg. N+60 Kg. P_2O_5 + 40 Kg. K_2O per hectare, whereas the highest disease intensity was recorded, when 120 Kg. N per hectare was given alone. No work appears to have been done on the host nutrition in respect of the incidence of Leaf spot of Dolichos bean caused by *A. alternata* before, but more or less similar effect of NPK on disease severity was obtained by Prabhu (1965), Ojha and Mehta

(1970) and Ram and Joshi (1981). They also observed that the application of nitrogenous fertilizers increased the susceptibility of Wheat plants to *A. triticina*. Prasad (1983), was also of the opinion that the incidence of Root and Stem Rot of Jute was inversely proportional to potassium levels and these results fully support the results of presents findings. In case of interaction of NPK on *Alternaria* leaf spot and Fruit rot of Brinjal caused by *A. alternata*. Singh (1988), observed primitive correlation of disease with increasing levels of Nitrogen, while phosphorus and potash, were responsible to decrease the incidence.

Attempts have been made to work out the biochemical basis of disease resistance, which is gaining significance at present in view of minimizing the pollution of biosphere and preservation of ecological balance endangered by indiscriminate use of chemicals for different biochemical parameters concerned to the resistance of leaf spot of Dolichos bean caused by *Alternaria alternata*. The leaves of diseased and healthy plants were analysed and it was observed that wax content was considerably reduced in 40 and 70 days after inoculation. Necrotic tissues were found to contain 12% of total wax present in the diseased leaves. The findings are in accordance with observations recorded by Bhaskaran and Kandaswamy (1977), who observed reduction in photosynthetic pigments (chlorophyll a and b) in the necrotic halo and pre-halo tissue of Sunflower leaves infected by *Alternaria helianthi*. The reduction in chlorophyll content may be due to decreased synthesis of these pigments in the infected tissue or destruction of a part of chlorophyll due to infection (Diener, 1963) and proteolysis causing degeneration of chloroplasts (Mayer *et al.* 1960). Reduction in chlorophyll content takes place due to inhibition of its synthesis by the fungus (Pero and Main, 1970) or may be due to reduction in number and size of chloroplast (Tu and Ford, 1968). Infected Cotton leaves due to *A. macrospora* were found to reduce chlorophyll a and b content by Padmanaban and Narayanaswamy (1978). Kumar and Rao (1980), observed the reduction in chlorophyll content in resistant and susceptible varieties of Wheat leaves infected with *A.*

trititcina.

The polyphenol content in healthy leaves, was recorded higher, which reduced successively 40 and 70 days after inoculation and only small quantity was present in necrotic tissues. Similar observations were observed by Main (1971), who found decrease in phenols towards the lesion centre infected with *A. alternata*, on Tobacco leaves. Kumar and Rao (1980), reported that phenol content was decreased in susceptible leaves of Wheat after inoculation with *A. trititcina*. Reduction in phenol of mandrin infected with *A. citri*, was also studied by Agrawal and Khara (1983). Chahal (1986) also found that young leaves of Brown Sarson having higher amount of phenols may feasible for their greater resistance to *A. brassicae* in comparison to olders.

The highest sugar content was found in healthy leaves and reduction after 40 and 70 days of inoculation and necrotic tissue. The reducing sugars were present in higher proportion than non-reducing sugars. Reduction in reducing and non-reducing sugars is due to presence of greater proportion of their nutrients present in host plants, which is utilized by the pathogen (Padmanaban, 1973 and Vyas and Panwar, 1976). These findings are in with the results of Bhaskaran and Kandaswamy (1977), who found that total soluble sugars, glucose and fructose decreased in the necrotic and halo regions. Chopra and Jhooty (1974), observed decrease in sucrose, raffinose and lactose in the necrotic area of leaf spot disease of Watermelon caused by *A. cucumerina*. The reduction in sugar content may be a result of degradative metabolism in diseased tissue (Nema, 1983). Since the physical presence of pathogen in leaf spot disease of Cotton, was confined upto necrotic area, in which decreased sugar content in this zone indicated the utilization by the pathogen (Bhaskaran and Kandaswamy 1977). Sugars of Barley leaves infected by *A. alternata* decline the disease development (Dixit and Gupta, 1983). Chahal (1986), found that young leaves contained higher amount of sugars accounting for their higher resistance to *A. brassicae* in

Brown sarson.

The nitrogen, phosphorus and potash contents, were found higher in healthy leaves and were reduced after 40 and 70 days of inoculation. The reduction of these contents was more pronounced in necrotic tissue. A greater proportion of these nutrients lying in host plant is utilized by the pathogen during pathogenesis. These findings are in accord with the observations of Khatri *et al.* (1986), who found that nitrogen, phosphorus and potash contents, were decreased in blighted (*A. porri*) halo and apparently healthy Onion leaf tissue present in close proximity of diseased tissue in comparison to healthy tissues. Pillayarsamy *et al.* (1973), found that the content of potash was drastically reduced in Chilly fruits infected with *A. solani*. Contrary to these results, Bhaskaran *et al.* (1975) and Kumar and Rao (1980) reported that total and amino-nitrogen increased in the susceptible varieties after inoculation with *A. macrospora* in Cotton leaves and *A. triticina* in Wheat respectively.

Reduction in sulphur content was found slightly reduced in necrotic tissue in comparison to healthy indicating that the pathogen requires only very small of sulphur for its development.

In studying role of seeds in the survival of pathogen and disease perpetuation, it was observed that fungus could be isolated from both naturally infected as well as artificially infested seeds from November to June till the next crop season. The seeds, when sown in the following Kharif season gave rise to seedlings with typical symptoms of *Alternaria alternata* to varied extents. Thus it was established that infected seeds were the carriers of the pathogen and served as a primary source of infection. The seed borne nature of this pathogen was established by other workers also. Higgins (1947), Wheeler (1958), Kapoor and Hingorani (1958), Bhowmik (1969), Desai (1971) and Gamawat and Prasad (1972), Agarwala and Singh (1974), Padaganur (1979), Narain and Saksena (1981), Allen *et al.* (1983), Singh and Suhag (1983), Prasad and Singh (1983), Naryanappa (1982),

Jeffrey *et al.* (1984), Kumar and Patnaik (1985), Raut (1985), Vijaylakshmi and Rao (1986), Simay (1987) and Krishnappa and Shetty (1990). Mustard seeds infected with *A. brassicae*, served as a source of primary inoculum as observed by Ansari *et al.* (1989). Patel and Desai (1971) and Ghamawat and Prasad (1972), reported that seeds infected by *Alternaria burnsii*, play an important role in the perpetuation of the disease and survival of the pathogen of Leaf blight of Cumin. The results of all these workers are in agreement with the present findings. Zazzerini and Buonauro (1981), reported *A. carthami* and *A. alternata* on Safflower infected leaves and found that infection takes place through seeds.

Investigations regarding the role of infested soil and disease plant debris in the survival of pathogen and perpetuation of the disease revealed that the pathogen could survive in infested soil as well as in the diseased plant debris for eight months in infective stage till the next growing season and the plants raised in pots filled with infested soil served as a source of primary inoculum for the pathogen. Similar observations were recorded by Khasanov and Nazarov (1970) in *A. alternata*, where they proved it as a soil borne fungus. Prabhu and Prasad (1967), reported that *A. triticina*, remained viable for more than 20 months in diseased plant debris under the laboratory conditions. Patel and Desai (1971) and Gamawat and Prasad (1972), observed that plant debris played an important role in the survival of perpetuation of *Alternaria burnsii*. Shukla *et al.* (1978), reported that *A. alternata* causing Stem blight of wheat was isolated from the underlying soil and plants grown in pots, containing contaminated soil, expressed disease symptoms. These results are fully in accordance with the present findings. Prasad and Reddy (1986) found that *A. alternata* was able to survive for three months on infected leaves exposed to natural conditions on soil surface and upto eight months on leaves kept in laboratory. Singh (1987), also reported that infested soil and plant debris served as a source of primary infection of *A. alternata* causing Leaf spot and Fruit rot of Brinjal.

The role of infested soil and diseased plant debris in the survival

and perpetuation of different species of *Alternaria*, affecting various crops were recorded also by Ghamawat and Prasad (1972), Allen *et al.* (1983), Jeffrey *et al.* (1984), Singh (1987), Ansari *et al.* (1989) and Tripathi (1992).

In studies on secondary spread of the disease through air, it was observed that healthy plants kept in vicinity of diseased plants exhibited typical symptoms of the disease by the air transmission of spores, thereby confirming the role of air in the spread of disease. The lower leaves were found to be infected for the first time. These findings are in consonance with the findings of Ghamawat and Prasad (1972), who reported in leaf of Cumin incited by *A. burnsii*, the secondary spread of the disease took place through aerial infection. The secondary spread of air borne conidia of *A. triticina*, *A. cucumerina*, *A. alternata* and *A. brassicae*, was also recorded by Ibrahim *et al.* (1975), Shukla (1975), Kumar and Arya (1976), Singh (1987) and Ansari *et al.* (1989) respectively. The observations of Shukla (1987), regarding the role of air in spread of Alternariosis is due to *A. brassicae* and *A. alternata* causing Blight of oil seed Crucifers and Leaf spot and fruit rot of Brinjal respectively also corroborate the present findings.

Investigations were carried out to locate the sources of resistance against *A. alternata* by subjecting 93 germplasms/cultures of Dolichos bean under natural conditions. Out of these six germplasms/cultures viz; Arka Vijai, Cultures viz; 7703, 7008-B and 9101, JDL-37 and Todi-125-136 were found to be tolerant being disease free (F). Five germplasms/cultures viz; Attapati, Culture-7022, JDL-17, Pusa Early Prolific and Rajani were found resistant (R). Twelve germplasms/cultures viz; Arka Jai, Cultures viz; 6802, 7001, 7103, 7301, 8001, 8002, 8403, HA-3, HD-4, HD-81 and JDL-85, were found moderately resistant (MR) and rest of the germplasms were found to be Moderately Susceptible (MS), Susceptible (S) and Highly Susceptible (HS) and under artificial epiphytotic conditions germplasms/cultures which were found diseased free (F), Resistant (R) and Moderately Resistant (MR) under natural conditions, ten germplasms/cultures viz. Arka Vijai, Cultures-6802,

7022, 8002 and 9001, HA-3, HD-81, Rajani and Todi-125-126, proved Moderately Resistant (MR) and rest of the germplasms, were found Moderately Susceptible (MS) and Susceptible (S), by inoculation of seeds and nine germplasms/cultures viz; Arka Jai, Cultures-7001, 7022 and 9101, DDL-37, HD-4, JDL-85, Rajani and Todi-135-136, were found Moderately Resistant (MR) and rest of the germplasms/cultures, were found Moderately Susceptible (MS) and Susceptible (S) by inoculation of potted plants. Various degrees of resistance and susceptibility in Sunflower germplasms/cultures, were reported by other workers. Gupta *et al.* 1978, Singh *et al.* 1984, Ghemawat *et al.* 1989, Shetty, 1990 and Sahu *et al.* (1991), who fully supported the present findings.

The host range studies of the pathogen was carried out using 70 plant species, belonging to 19 families, under artificial conditions of inoculation, it was observed that 50 plant species including *Abelmoschus esculentus*, *Abutilon indicum*, *Althea rosea*, *Allium cepa*, *Argemone mexicana*, *Avena sativa*, *Arachis hypogea*, *Brassica campestris*, *B. campestris* var. *dichotoma*, *B. juncea*, *B. oleracea* var. *botrytis*, *B. oleracea*, var. *capitata*, *B. oleracea* var. *gongylodes*, *Carrisa carandus*, *Cajanus cajan*, *Capsicum annuum*, *Carthamus tinctorius*, *Chenopodium album*, *Chrysanthemum indicum*, *Colocasia antiquorum*, *Coriandrum sativum*, *Crotolaria juncea*, *Cucurbita maxima*, *Cynodon dactylon*, *Dahlia* sp., *Datura alba*, *Gossypium* sp., *Glycine max*, *Hibiscus rosa-sinensis*, *Hordeum vulgare*, *Lagenaria vulgaris*, *Linum usitatissimum*, *Lycopersicum esculentum*, *Luffa cylindrica*, *Ocimum sanctum*, *Pennisetum typhoides*, *Pisum sativum*, *Raphanus sativus*, *Ricinus communis*, *Saccharum officinarum*, *Sesamum indicum*, *Sida acuta*, *Solanum nigrum*, *S. melongena*, *S. tuberosum*, *S. xanthocarpum*, *Sorghum vulgare*, *Tagetes erecta* and *Triticum aestivum*, the pathogen produced, more or less similar symptoms as observed in nature, thereby confirming that the pathogen has a wide host range. These findings are in accordance with the results of Young (1926), who reported that although *A. alternata*, appeared as a weak parasite on many plant species but *Brassica oleracea*, *B. rapa*,

Capsicum annuum, *Daucus carota*, *Hordeum vulgare* and *Solanum nigrum*, were its true hosts. Neergaard (1945), reported *A. alternata* as a pronounced polyphagous facultative parasite. Kapoor and Hingorani (1958), observed that the Brinjal isolated infected *Solanum tuberosum* L. and *Lycopersicon lycopersicum* L. also. The polyphagous nature of *A. alternata*, was subsequently confirmed by other workers (Singh and Tandon 1967 and Crisan 1976). In the present studies the pathogen proved its cosmopolitan nature by various workers namely Young (1926), Dhanraj (1970), Gupta (1970), Sokhi *et al.* 1972; Janardhan and Hussain (1972), Kapoor and Hingorani (1958), Narain and Saksena 1974, 1975a and 1975 b, Crisan 1976, Shukla and Bhargava, 1976, Patil *et al.* (1981), Narayanappa (1982), Prasad (1983), Singh and Suhag (1983), Singh *et al.* (1984), Khan *et al.* 1984; Prakash and Roof 1985 and Gupta *et al.* 1987.

Bio-assay test of 26 fungitoxicants belonging to different groups (Benzene, Copper compound, Dithiocarbamate, Heterocyclic nitrogen compound, Organomercurials, Quinone, Sulphur compound, Systemic and an Antibiotic viz; Agrosan G.N., Bavistin, Benlate, Blitox- 50, Brassicol, Calixin, Captan, Ceresan (Dry), Dichlone, Dithane M-45, Dithane Z-78, Duter, Emisan-6, Ferbam, Foltaf-80-W, Hexaferb, Karathane, Kavach, Pancotine, Ridomil, Spergon, Suflex, Thiram, Vitavax and Ziram and an antibiotic (Aureofungin) against *Alternaria alternata*, revealed that twelve of them namely Agrosan G.N., Aureofungin, Captan, Ceresan (Dry), Duter, Emisan- 6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax proved to be most effective as they inhibited the growth of the fungus completely. The rest of fungitoxicants, were categorised as partially effective amongst them. Dithane M-45 caused the highest inhibition of the fungal growth followed by Dichlone. The other fungicides in the descending order of efficacy were Suflex, Calixin, Blitox- 50, Brassicol, Ridomil, Benlate, Ferbam, Ziram, Spergon, Bavistin, Hexaferb and Dithane Z-78.

In hyphal dry weight method out of the 25 fungicides and an

antibiotic (Aureofungin) examined in hyphal dry weight method, Agrosan G.N., Aureofungin, Captan, Ceresan (Dry), Duter, Emisan- 6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax, proved to be most effective as they inhibited the growth of fungus. Calixin and Suflex inhibited the growth of fungus up to 13.70 mg. at 0.05 per cent, 10.70 mg. at 0.10 per cent; 10.90 mg. at 0.15 per cent; 10.25 mg. at 0.20 per cent; 8.35 mg. at 0.40 per cent and 4.30 mg. at 0.60 per cent and 10.97 mg. at 0.05 per cent; 7.54 mg at 0.10 per cent; 6.50 mg. at 0.15 per cent; 8.43 mg. at 0.20 per cent; 5.20 mg. at 0.40 per cent and 4.28 mg. at 0.60 per cent concentrations respectively. The remaining fungitoxicants viz; Bavistin, Benlate, Blitox-50, Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon and Ziram were found partially effective in checking the growth in various concentrations of fungitoxicants, with a significant variation in dry weight.

In lesion development test out of twenty five fungicides and an antibiotic (Aureofungin), tested ten of them namely Agrosan G.N., Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax as in hyphal dry weight method proved to be most effective as it did not produce any pathogenic effect. Other fungicides namely Brassicol, Calixin and Suflex, were found effective in producing Light yellow (LY) lesions only in different concentrations of fungitoxicants and proved better in comparison to remaining fungitoxicants tested except Brassicol in 0.30 per cent, produced no pathogenic effects. The Phytotoxic effects (PE) were noticed in the treatment of Dithane M-45 and Vitavax, while Extensive Brown (EB) spots were recorded in the treatment with Bavistin, Dithane Z-78 and Ziram in different concentrations, except Light Brown (LB) lesions in 0.10 per cent concentrations in treatment with Bavistin. Other fungitoxicants viz; Dichlone, Ferbam, Ridomil and Spergon firstly produced Extensive Brown (EB) spots in lower concentration of 0.05 per cent and thereafter in 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.25 per cent and 0.30 per cent (higher concentrations),

decreased the intensity of pathogenic effect exhibiting Light Brown (LB) lesions. The remaining fungitoxicants viz; Benlate, Blitox-50 and Hexaferb shown Yellowish Brown (YB) lesions in different concentrations except Benlate and Hexaferb, which exhibited Light Yellow (LY) lesions in 0.05 per cent and 0.30 per cent concentrations. In the test Phytotoxic effects (PE), were found decreased with increase in concentration.

In spore germination test (spore germination, number of germ tubes and length of germ tube), out of the twenty four fungicides and an antibiotic (Aureofungin), Agrosan G.N. Aureofungin, Captan, Ceresan (Dry); Duter; Emisan-6, Foltaf 80-W, Karathane, Pancotine, Thiram and Vitavax proved to be most effective as spores failed to germinate. The other fungitoxicants viz; Dichlone and Dithane M-45 produced minimum spore germination being less effective and proved better in comparison to other fungitoxicants. The inhibition in spore germination over control was found 72.04 per cent and 79.56 per cent in 0.0002 per cent; 78.57 per cent and 79.56 per cent in 0.0004 per cent and 81.91 per cent and 84.04 per cent in 0.0006 per cent concentrations. The remaining fungicides viz; Bavistin, Benlate, Blitox-50, Brassicol, Calixin, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram, were found particularly effective in arresting the spore germination. The significant variations in inhibition over control, were also recorded ranging from 15.75 per cent to 81.81 per cent in different concentrations of fungitoxicants tested in respect of germination of seeds.

The number of germtubes varied from 6-37, 5-30 and 2 to 29 in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants. Minimum 6, 5 and 2 germ tubes were produced in treatment with Dithane M-45 in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations respectively, while the maximum number of germ tubes 37, 30 as well as 29 in the treatment with Calixin in 0.0002, 0.0004 and 0.0006 per cent concentrations respectively, were observed. Other fungitoxicants viz;

Bavistin, Benlate, Blitox-50, Brassicol, Dichlone, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram produced 10, 28, 36, 32, 11, 12, 28, 36, 32, 11, 12, 28, 16, 30, 20, 27 and 20 in 0.0002 per cent concentration, whereas 18, 23, 27, 28, 9, 11, 20, 14, 25, 18, 28 and 17 germs tubes were found respectively in 0.0004 per cent concentration of Bavistin, Denlate, Blitox-50, Brassicol, Dichlone, Dithane Z-78, Ferbam, Hexaferb, Ridomil Spergon, Suffex and Ziram respectively. In 0.0006 per cent concentratioin of Bavistin, Benlate, Blitox-50, Brassicol, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram produced 6, 28, 27, 27, 21, 3, 16, 11, 25, 18, 28 and 14 germtubes respectively.

As far as the length of germ tubes is concerned the minimum germ tube length measuring 0.0005 mm; 0.0007 mm. and 0.006 mm. were recorded in the treatment with Bavistin in 0.0002, 0.0004 and 0.0006 per cent concentrations of fungitoxicant respectively, whereas maximum number of germ tubes measuring 0.017 mm., 0.014 mm. and 0.013 mm. with treatment to Calixin in 0.0002, 0.0004 and 0.0006 per cent concentrations of fungitoxicants respectively. Other fungitoxicants namely, Benlate, Blitox-50, Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram produced germ tubes measuring 0.012 mm; 0.017 mm; 0.018 mm; 0.008 mm; 0.006 mm; 0.006 mm; 0.006 mm; 0.009 mm; 0.013 mm; 0.012 mm; 0.015 mm; 0.013 mm; 0.012 mm; 0.015 mm; 0.013 mm; 0.017 mm. and 0.018 mm. in length respectively in 0.0002 per cent concentration, while 0.015, 0.016, 0.015, 0.008, 0.008, 0.008, 0.012, 0.008, 0.013 m.m., 0.007, 0.014 and 0.012 mm. were recorded in 0.0004 per cent concentration of Benlate, Blitox-50 Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexaferb, Ridomil, Spergon, Suflex and Ziram respectively. In 0.0006 per cent concentrations of Benlate, Blitox-50, Brassicol, Dichlone, Dithane M-45, Dithane Z-78, Ferbam, Hexferb, Ridomil, Spergon, Suflex and Ziram produced germtube measuring length of 0.009 mm.; 0.016 mm.; 0.013 mm.; 0.012 mm.; 0.013 mm.; 0.007 mm.; 0.012 mm.; 0.008 mm.; 0.008 mm.; 0.009 mm.; 0.010 mm.; 0.008 mm.; 0.004 mm. and 0.009 mm. respectively.

The inhibition in all the three parameters in spore germination, germ tube length and number of germ tubes produced, were pronounced in higher concentrations of fungitoxicants with significant differences between different fungitoxicants and their concentrations, Dichlone and Dithane M-45 caused appreciable degree of inhibition in spore germination, germ tube number and germ tube length. The germ tube length was inhibited nearly 78.19 per cent at higher concentrations of fungicides. At lower concentration, Blitox-50 and Calixin, were not found effective but ineffective at higher concentrations of the inhibition of spore germination.

The reports of inhibition of mycelial growth of *Alternaria alternata* in *vitro* by Captan, Agrosan, Thiram and Zineb, were made by Misra and Singh (1965), Sahni and Singh (1967) and Singh and Malika (1974), which corroborate the present findings, while Misra *et al.* (1970), observed Thiram and Zineb to be the most effective against this fungus. The best control of the disease was achieved with Captan, Ferbam and Maneb at 0.5 per cent to 0.025 per cent by Crisan *et al.* (1970). Duter and Cuman, completely checked the growth of *A. alternata* (Chauhan, 1970). Ramkrishnan *et al.* (1971), found 0.1 per cent Dithane Z-78 and 0.20 per cent Duter as more effective. Singh and Malika (1974), in their studies reported Captafol, Dithane M-45 and Thiram to be most promising chemicals against *A. alternata*. Crisan (1976), found Vitavax to be effective against all the isolations of *A. alternata* under study. The efficacy of seventeen fungicides was also studied on the spores of *Alternaria alternata* of Cotton (Yousef *et al.* 1977). Ziram was found most suitable fungicide in *vitro* test against *Alternaria helianthi* (Bhaskaran and Kandaswamy, 1977 and Mukewar and Gera 1980). Aureofungin and Brestanol, were noticed to be effective against *A. alternata* in laboratory test as reported by Mathur and Sarbhoy (1977 and 1983). Thiram was concluded to be best fungicide as recorded appreciable degrees in growth rate and spore germination of *A. alternata* causing leaf spot of Soyabean followed by Captan and Verdesan (Kuthubutheen and Pugh, 1978). Narain (1978), found

Difolatan, Plantavax, Benlate, Bavistin, Dithane M-45, Dithane Z-78 and Duter to arrest completely the growth of *A. alternata* in *vitro*.

Utikar *et al.* (1979) and Crisan *et al.* (1979) reported Zineb and Dithane M-45 to be more superior to other fungicides against the fungus, *A. alternata*. Mancozeb dithiocarbamate, was proved to be most effective in controlling the disease 17.0 per cent to 33.0 per cent and yield increase between 43.0 - 65.0 per cent (Kolte *et al.* 1979), whereas Thiram and Duter followed by Aureofungin and Captan, were found to be effective as reported by Basavarajaiah *et al.* (1979).

Pancotine, was found most effective against *A. macrospora* the incitant of leaf spot disease of Cotton in *vitro*, followed by Cuman, Dithane M-45, Duter and Blitox as observed by Desai and Patil (1982). Kalra and Sohi (1984), found that except Calixin all the systemic and non-systemic fungicides including Thiram, Dithane M-45 and Captafol completely inhibited the growth of *A. alternata*. Siddaramaiah and Desai (1984), found Dithane M-45 (Mancozeb) and Pancotine as most inhibitory to growth of *A. macrospora* from Cotton. Zineb, was found to be most suitable fungicide in exhibiting morphological changes in *A. alternata* and *A. helianthi* colonies and controlling the fungus in *vitro* followed by Captan (Ungaro and Azevedo, 1984). Mali and Joi (1985), found that Difolatan (Captafol), Thiram and Vitavax, proved to be most effective against colony growth and sporulation of *Alternaria alternata*. Sharma and Chauhan (1985), recorded that spore germination of *Alternaria macrospora*, *Curvularia lunata*, *Helminthosporium spiciferum* and *Myrothecium roridum*, was completely inhibited by Blitox (Copper oxychloride), Dithane Z-78 (Zineb) and Difolatan (Captafol) at 100 and 500 ppm. and Dithane M-45 and Syllit (Dodine) at 500 ppm. Vasu and Roy (1985), found that 3, 4, 5- trichlorophenyl phosphate proved to be most effective against *Alternaria alternata* in *vitro*. The effectiveness of fungicides in aerial spraying of Sunflowers to control erogenic infection, was studied by Kronenberg *et al.* (1991). The results are in consonance with the present

findings.

The twelve seed dressing fungitoxicants, which proved effective in laboratory screening, were selected to assess their efficacy in seed germination and seed borne infection of pathogen *in vitro*. These studies revealed that all the seed dressing fungitoxicants improved seed germination. The maximum seed germination 96.49 per cent was recorded in case of seed treatment with Captan, followed by 92.65 per cent in Ceresan (Dry). Lowest seed germination 64.28 per cent was recorded in treatment with Vitavax. It was also recorded that seeds treated with Ceresan (Dry) had no infection. The lowest infection 1.29 per cent was recorded in treatment with Captan, whereas maximum 14.55 per cent infection was recorded in the treatment with Vitavax, followed by Pancotine, Thiram, Emisan-6, Duter, Aureofungin (Antibiotic), Agrosan G.N., Foltaf 80-W, Karathane, Kavach and Captan ranging from 13.83 per cent, 12.79 per cent, 11.91 per cent, 10.26 per cent, 5.85 per cent, 2.83 per cent, 2.20, 1.65 per cent, 1.34 per cent and 1.29 per cent respectively. In general all the fungitoxicants examined were found significant over control in reducing the seed borne infection.

In Pot experiment for ascertaining the efficacy of eleven fungitoxicants on seed germination and control of seedling infection, it was found that significant increase in seed germination 95.47 per cent resulted by seed treatment with Ceresan (Dry), only followed by 92.08 per cent with Captan, although numerical increase in seed germination over control was given by other fungicides viz; Thiram, Agrosan G.N., Emisan-6, Duter, Pancotine, Aureofungin, Karathane, Kavach and Vitavax. Poor germination 77.34 per cent was recorded in Foltal-80W. It was further noted that Ceresan (Dry) Thiram and Vitavax controlled the seed borne infection of *A. alternata* completely. Significant control of the disease was also given by Captan and Agrosan G.N. but other fungitoxicants viz; Aureofungin (Antibiotic), Duter, Emisan-6, Karathane, Kavach, Pancotine and Foltaf 80-W, gave poor results in reducing seedling infection up to 4.39 per cent, 10.75 per cent, 18.85 per

cent, 13.58 per cent; 12.96 per cent, 12.94 per cent and 20.30 per cent as compared to control. These findings are in consonance with the observations of Gentner (1923), who found reduced infection of *Alternaria* sp. in seeds treated with Ceresan, Mercurian and Thiram. Black (1946), noticed to protect seed germination up to 3-5 years by treating the seeds with Ceresan wet or dry stored at 53°C - 75°C and 50.0 per cent relative humidity. The findings are also in agreement with Dharmvir (1972).

Increase in germination percentage in the seeds treated by Mills (1975). Jhamaria *et al.* (1974). Kumar and Urs (1976), Narain (1978) and Prasad and Das (1986), found Captan, Agrosan G.N. and Ceresan to be most effective fungicides respectively in eliminating the seed borne infection of *A. alternata* isolated. Srivastava and Saksena (1974) and Lokesh *et al.* (1987), reported the control of seed borne infection of *Alternaria* sp. affecting the seeds of various crops. Suhag (1975), during the course of his studies with regards to improvement of seed germination by treatment with Agallol, Ceresan, Captan and Thiram. Seed Treatment with fungicide, Fytolan gave the highest yield as recorded by Bhaskaran and Kandaswamy (1977). Shukla *et al.* (1980), reported that Agrosan G.N., Thiram, Benlate and Dithane M-45 controlled the seed borne infection of *A. alternata* on triticales seeds most effectively.

Thakur *et al.* (1981), observed significant pre and post-emergence losses in Radish seeds infected with *A. alternata*, *Cladosporium cladosporioides* and *Aspergillus niger* and found 0.20 per cent Agrosan G.N. and Ceresan (Dry) to give effective control of seed borne fungi. Eleven seed dressing fungicides, were examined by Vishunavat and Shukla (1982), against the fungi associated with Lentil seeds and found that Captan eliminated all the fungi including *Alternaria*. Prasad (1983), found that seeds treated with Dithane M-45 (Mancozeb) exhibited high germination percentage. Jain *et al.* (1983), found similar results in seed treatment with Bavistin and Captan as best control of Pre and post-emergence seed rot caused by *A. alternata* and

Curvularia lunata. Later Gupta *et al.* (1984), recommended Agrosan G.N., Dithane Z-78 and Dithane M-45 as seed protectants for Pigeonpea. Seed treatment with Bavistin and Difolatan 80-W (Captafol), was found effective in eradicating the pathogen, *Alternaria alternata* and improving germination by 8.50 per cent to 24.50 per cent. *Alternaria alternata* invading 78.0 per cent seed samples of Sunflower were effectively checked by Aureofungin, Blitox and Vitavax. Benlate was less effective as observed by (Vijayalakshmi and Rao, 1985). Kumar and Patnaik (1985), during the study of fungicidal treatment of *Alternaria alternata* infected with Pigeon Pea, seven fungicides significantly improved seed germination, seedling, root length, shoot length and dry weight. Mali and Joi (1985), found Captafol, Thiram and Vitavax as most effective seed dressing fungicides against *A. alternata*. Shrotri *et al.* (1985), recorded that *A. alternata* invading Dahalia seed was effectively controlled by the application of Ceresan. Most effective treatment of seeds by soaking for 1-2-hours in Ceresan or Duter, was concluded by Padganur and Basvaraj (1987) Kumar and Singh (1996), reported that Emisan-6, Bavistin and Bavistin 25-SD (each 219 kg.) treated seeds controlled *A. alternata* with seed of winged bean and also increased seed germination. The observations of all these contributions are in accordance with the present findings.

In the present study the efficacy of twelve spray fungitoxicants found completely or partially effective in bio-assay test was evaluated in Pot-experiments. Out of these Captan gave the best control of disease as well as increase in yield followed by Thiram. The remaining fungitoxicants in order of descending merit, were Ceresan (Dry), Karathane, Kavach, Agrosan G.N., Duter, Aureofungin (Antibiotic), Emisan-6, Pancotine, Foltaf 80-W and Vitavax proved poorest amongst all the fungitoxicants tested but gave significantly better yield followed by Foltaf 80-W, which exhibited lowest yield weighing 19.94 gms. and 20.35 gms. and 19.62 gms. and 19.82 gms. 2001 and 2002 respectively. These findings are in consonance with the observations of Singh and Singh (1971), who observed that Dithane M-45 (0.25 per cent) and Dithane Z-78 (0.25 per cent) effectively controlled the

Alternaria blight of Wheat with corresponding increase in yield. In 1980, Mansour reported the leaf spot. (*A. alternata*) of Faba beans, was effectively controlled by four sprays of Plantavax and Dithane M-45 (each 250/1000 liter water), resulting increase in yield. Additionally Singh and Shukla (1984), Singh and Sharma (1986) and Kumar and Pandey *et al.* (1987), found that Dithane M-45 was the most effective fungitoxicant against *A. alternata* causing leaf spot and Fruit rot of Brinjal, Alternaria Fruit rot of Tomato and Leaf Spot of Onion. Maheswari *et al.* (1991), examined the efficacy of six fungicides in the field trial during 1989-1991. Out of which the most effective control (64.70 per cent) of *A. solani*, causing Early blight of Tomato, was given by Copper oxychloride, followed by Mancozeb (61.70 per cent). The report of this work fully supported the present findings. Solanki *et al.* (1997), tested the efficacy of fungicides and found Thiram and Carbendazin BDN2 to be best for promoting higher germination.

In field evaluation same set of fungitoxicants, was also carried during 2001 and 2002, for the control of Leaf spot of Dolichos bean. It was observed that the best control of the disease with corresponding increase in yield was obtained by the application of Captan followed by Thiram, which were different from each other. The performance of other fungitoxicants was comparatively poor, which in descending order of efficacy were Ceresan (Dry), Karathane, Kavach, Duter, Agrosan G.N., Aureofungin (Antibiotic), Emisan-6, Pancotine and Ceresan (Dry), Agrosan G.N., Aereofungin (Antibiotic), Duter, Emisan-6, Pancotine, Foltaf-80-W and Karathane, differed significantly in the years 2001 and 2002 respectively. Foltaf-80W fungitoxicant, was found poorest amongst all the fungitoxicants tested controlling the disease in both the years 2001 and 2002. The present results are in corroboration with the findings of Sindhamathan *et al.* (1976), where they reported that Alternaria Leaf spot of Sunflower caused by *A. helianthi*, was controlled by spraying Dithane M-45. Subsequently Narain (1978), reported that amongst the spray fungicides Captafol and Dithane M-45 followed by Zineb, proved to be the best fungicides in controlling leaf spot of Sunflower caused by *A. alternata*,

Two applications of Dithane M-45 or Dithane-Z-78, were reported to be the most effective in controlling the Alternaria Leaf spot of wheat incited by *A. alternata* (Agrawal *et al.* 1976). Fungicides like Brestan 60, Dithane Z-78, Dithane M-45, Dithane Z-78 and Captafol, had significant impact in controlling different types of diseases caused by *Alternaria* sp. (Gupta *et al.* 1977; Sharma 1986 Shivpuri *et al.* 1988; Vishwakarma 1990; Reddy *et al.* 1991 and Tripathi, 1992). Fungicidal management of Leaf spot of Sunflower incited by *Alternaria helianthi* was recorded by Patel *et al.* (1995), Vishwakarma and Pandey (1995) and Khan *et al.* (1999). The findings recorded proved superior over control.

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Chapter - VI
SUMMARY

SUMMARY

Dolichos bean (*Dolichos lablab*, L), is one of the significant vegetable yielding crop of India and other countries of the world but Pakistan, Brazil, China, Egypt, Mexico, Sudan, U.S.A. and U.S.S.R. account for nearly 65.0 of the total production and having ample potential to tide over the paucity of vegetable growing in the country due to a rich source fo protein, minerals, vitamins and enzymes and enriches in the soil due to fixation of atmospheric nitrogen by root module bacteria as well as used in preventing soil erosion. Dolichos bean occupies a premier place in the national economy of our country and prosperity of millions of countries, rural and urban populations, who depends upon harvest of vegetable yielding cash crop. Inspite of the presence of improved varieties and large acreage under its cultivation the total production in India is very low in comparison of other countries of the world, mainly due to off-setting of the foliar diseases of fungal origin as well as bacterial, viral and nematodal diseases. Among these, the leaf spot of Dolichos bean caused by *Alternaria alternata* (Fries.), Keissler, gradually increasing on new evolved high yielding varieties of Dolichos bean, has been recorded to be predominated in Uttar Pradesh during recent years, about which, there is no information available, requiring the immediate attention of Plant Pathologists, if the production of this crop is to be boosted in the country. So far, with a view to combat this serious malady effectively, the present investigation was taken to find out the prevalence and severity of disease in U.P., to investigate the morphological and cultural characters of the fungus, to ascertain the role of enzymes in pathogenesis in *vivo* and *vitro*, effect of pathogen on biochemical constituents of diseased parts of the host, growth and sporulation on different carbon sources and nitrogen inculture, effect of different doses of nitrogen, phosphorus and potash on the severity of disease, susceptible growth period of host, influence of climatic conditions on the development of disease, host range relationship of the pathogen, disease perpetuation as well as source of resistance, with an aim

of its control managing the strategy of disease, and the results sought are discussed here in brief.

Leaf spot of Dolichos bean caused by *Alternaria alternata* (Fries.) Keissler, was found to be moderate to heavy in severity widely prevalent under different agroclimatic conditions of Uttar Pradesh as evident from the survey conducted during the years, 2001 and 2002 in thirty localities during Kharif season. The incidence of disease at different research stations during Kharif seasons of the year 2001 and 2002 varied from 13.48 per cent to 31.57 per cent from the germplasms/cultures viz., Culture-7301, Kamgranj Selection-2, Culture-9012, Culture-7015, Culture-9118, Culture-7708, HD-4, Arka Jai, DB-1, Culture-8403, Todi-125-136, Culture-7604, Pusa Early Prolific, Goyal, JDL-79, Rajani, Culture-6801, Culture-8101, Culture-7710, Hatikan, HD-66, Culture-7210, Culture-9109, Culture-9113, Culture-8005, TDL-85, HD-93 and Kalyanpur Type - 1 and 10.25 per cent to 34.85 per cent from the germplasms/cultures viz; Culture-7301, Culture-7210, Akra Jai, Culture-9118, Culture-6801, Hatikan, Culture-8005, Culture-7015, Todi-125-126, Goya, JDL-85, Pusa Early Prolific, Culture-9102, Kamgranj Selection - 2, HD-4, Culture-9109, Culture-8101, HD-66, DPL-1, Culture-8403, DB-1, HD-93, Rajani, JDL-79, Culture-7604, Culture-7710, Culture-9113, Culture-9104 and Kalyanpur Type - 1 respectively. The maximum disease incidence 31.57 per cent and 34.85 per cent was recorded at Vegetable Research Farm, Chandra Shekhar Azad University of Agriculture and Technology, Kanpur in both the years from the germplasm/culture, "Kalyanpur Type - 1", followed by 30.80 per cent and 29.26 per cent at Government Agriculture Centre, Jalaun and Crop Research Farm, Modipuram Area, Meerut from the germplasms/cultures HD-93 and Culture-9113 and rest of locations, while minimum 13.48 per cent and 10.25 per cent disease incidence, was recorded at Agriculture Science Centre, Ganiwa, Chitrakoot in both the years.

The average disease incidence ranged from 13.48 per cent to 31.57 per cent and 10.25 to 34.85 per cent in both the years of survey at different crop Research Stations.

Disease intensity during Kharif season varied from 22.40 per cent to 38.40 per cent from the germplasms / cultures viz; Todi-125-126, JDL-79, Culture-9113, Culture-7210, Akra Jai, Culture-6801, Hatikan, Culture-9102, Culture-7301, Culture-7708, Goya, Culture-8005, Culture-9118, HD-66, Culture-7710, Culture-8403, DB-1, HD-4, Pusa Early Prolific, Kamgranj Selection-2, HD-93, Culture-7015, Culture-8101, Rajani and Kalyanpur Type-1 and 17.90 - 40.50 per cent from the germplasms / cultures viz; Culture-7710, Culture-9118, Culture-9102, Hatikan, Culture-9104, Akra Jai, Culture-7301, Culture-7604, Culture-9109, DPL-1, Culture-8005, Kamgranj Selection-2, Goya, Culture-6801, HD-93, Pusa Early Prolific, JDL-79, Rajani, Culture-8403, Culture-7015, DB-1, HD-4, JDL-85, HD-66, Culture-9113, Todi-135-136, Culture-7210, Culture-8101 and Kalyanpur Type-1 in the years 2001 and 2002 showing wide spread prevalence in nature. Maximum 38.40 per cent and 40.50 per cent disease intensity was recorded from the germplasm / culture, "Kalyanpur Type-1" at Chandra Shekhar Azad University of Agriculture and Technology, Kanpur followed by 37.80 per cent and 39.50 per cent from Oil Seed Research Farm, Kalyanpur, Kanpur and Crop Research Farm Saraimira, Farrukhabad from the germplasms/cultures 7604 and 8101 and rest of locations in both the years, 2001 and 2002, while minimum 22.40 per cent and 17.90 per cent disease intensity was recorded from Directorate of Vegetable Research Farm, Varanasi and Government Agriculture Centre, Atarra, Banda from the germplasm / cultures Todi-125-136 and Culture-7710 in both the years respectively.

In general the maximum disease intensity was recorded from Chandra Shekhar Azad University of Agriculture and Technology Kanpur.

The disease incidence as well as disease intensity could not be recorded in the years 2001 and 2002 at Crop Research Farm, Deegh, Kanpur and Regional Agriculture Research Station Dilip Nagar, Kanpur and Regional Centre, Amroha, Jhansi respectively.

The investigations on the symptomatology of the disease, revealed that

under natural conditions, the disease appeared in the month of December and January in Kharif season crop and symptoms were confined on the upper surface of leaves only. The spots observed as small; brown to black; circular to oval with paler margins and Yellow halo measuring 0.15-1.0 cm. in size with characteristic concentric rings and cracked centre. The lesions at first were recorded as smaller in size, while in later stage the spots were found numerous extending over the whole leaf surface due to coalescence of adjacent spots, which later became perforated due to falling away of dead tissue.

According to Koch's postulates the pathogen, was isolated on two per cent Potato dextrose agar medium by transferring surface sterilized portions of diseased leaf and subsequently, it was purified by Single Spore Culture technique. In order to test pathogenicity reisolations of the fungus, were done and the results indicated that all the isolates of fungus proved pathogenic on Dolichos bean plants. The fungus was able to cause leaf spot disease even inoculation was done without injury, thereby indicating that it was pathogenic. The infection percentage 87.50 per cent was recorded on injured leaves in comparison to un-injured leaves, which showed 35.0 infection percentage.

Different methods of inoculation proved that the disease was always more in case of pin pick inoculation than without injury indicating the fact that injured leaves provided avenues for the pathogenic attack.

The morphological characters of the pathogen, were studied on Potato dextrose agar medium, revealed that colonies were moderately fast growing, which in the beginning dull; white; fluffy; circular and later turned into dark; greenish olive with abundant sporulation. Mycelium was found as septate; branched; hyaline; later turning into black and olive buff in colour measuring 3.20-8.60 μ in width. Conidiophores arise singly or in groups usually simple; septate; straight or bent; sometimes branched; swollend terminally; geniculate and dark olive buff measuring 24.50-69.30 \times 3.20 - 6.40

μ. Conidia, were formed in chains of 3-21; muriform; ovoid to obclavate; obpyriform; catenate (3-7); dark olive buff in colour; smooth sometimes verrucose with age; with 1-6 transverse septa and 0-6 longitudinal septa measuring $16.50-40.90 \times 8.20-12.50$ μ. Beaks were found usually short; light olive buff in colour and conical or cylindrical measuring $4.60-6.90 \times 3.40-5.80$ μ in size with 0-2 transverse septa. Chlamydospores were recorded sometimes terminal and intercalary and dark olive buff in colour, measuring $12.70 - 22.80$ μ in diameter. On the basis of morphological characters the fungus, under study has been identified as *Aternaria alternata* (Fries.), Keissler causing leaf spot of Dolichos bean.

The effect of different media viz; Non-synthetic semisynthetic and synthetic on the growth and sporulation of the fungus on, was studied and recorded the best growth on Potato Dextrose agar medium measuring 92.40 mm. followed by Richard's agar, Czapek's agar, Coon's agar, Leaf decoction agar, Malt salt agar, Sabouraud's agar, Corn meal agar and Asthana and Hawker's agar media. The least minimum growth measuring 17.46 mm. was obtained on Brown's agar medium.

Apart from studying the radial growth and sporulation of the pathogen various other cultural characters viz; growth; shape; zonation and colour of colony; substratum colour; pigmentation; colour of hyphae; colour of conidiophores; number and septation of conidia; variation in shape; size and colour existence and size of chlamydospores are quite different on the different types of solid media belonging to Natural (non-synthetic), Semi-synthetic and Synthetic media.

The results revealed that colony growth was found good; compact and raised; good compact with downy appearance; good sparse, thick and cottony; good sparse with entire margin; average sparse with entire margin; average sparse with suppressed hairy margin; average compact; good and semi-suppressed, compact; thin and cottony; average sparse with entire margin and poor sparse with entire margin on Potato dextrose agar, Czapek's

agar, Richard's agar; Leaf decoction agar; Coon's agar; Corn-meal agar; Malt salt agar; Sabouraud's agar; Asthana and Hawker's agar and Brown's agar media respectively. The colony shape was recorded almost circular on all the media except Czapek's agar medium, which exhibited lobe shaped colony. The colour of colony was almost recorded as dark grey; green with greenish tinge at the marginal ends; smoky grey; olivaceous black; dark black; dark greenish with darker centre; dark greenish with whitish margin; light green and creamy white on Asthana and Hawker's agar; Brown's agar; Sabouraud's agar; Malt salt agar, Richard's agar; Potato dextrose agar; Leaf decoction agar and Corn meal agar and Czapek's agar and Coon's agar media, respectively.

Substratum colour, was also found as irony grey; olive grey; light green, blackish green grey; olivaceous black; light vinaceous cinnamon; dark quaker drab and white smoky grey on Potato dextrose agar; Brown's agar; Asthana and Hawker's agar; Corn-meal agar; Czapek's agar; Leaf decoction agar; Malt extract agar; Richard's agar; Sabouraud's agar and Coon's agar media respectively. Zonation was found as distinct on Potato dextrose agar and Richard's agar media; not clear on Asthana and Hawker's agar medium; clear from upper side on Coon's agar and Czapek's agar media; clear from underside on Corn meal, Leaf decoction and Malt salt agar media, less clear from bottom side and absent from upper side on Brown's agar medium and dark quaker drab on Sabouraud's agar medium. Pigmentation was found absent on different types of media.

Colour of hyphae, was recorded as olive buff; mid olive; olive buff to greyish; pale olive buff; pale olive grey; light olive grey and colourless to greyish on Czapek's agar, Richard's agar and Sabouraud's agar media; Brown's agar; Malt salt agar; Asthana and Hawker's agar; Coon's agar; Corn meal agar; Leaf decoction agar and Potato dextrose agar media respectively. Hyphae were found septate varying in size from 2.50-8.0 μ in width on different types of media under study.

Colour of conidiophores, was recorded as olive buff to brown; midolive to brown; olivaceous brown; dark olive buff; pale to olive brown; dark olive brown and olive grey on Brown's agar and Richard's agar media, Czapek's agar and Sabouraud's agar media; Coon's agar and Malt salt agar media; Asthana and Hawker's agar medium, Potato dextrose agar medium, leaf decoction agar and Corn meal agar media respectively. The conidiophores varied in size from $24.20-68.0 \times 3.10-7.80 \mu$ on different types of media under study.

Conidia, were also found as olivaceous to dark brown; dark olive brown; olive buff to brown; light brown; dark olive grey and deep olive brown on Czapek's agar; Malt salt agar; Richard's agar and Sabouraud's agar media; Potato dextrose agar; Asthana and Hawker's agar; Brown's agar; Coon's agar; Corn meal agar and Leaf decoction agar media. Conidia were found borne in chains of 2-4 on Asthana and Hawker's agar, Brown's agar and Coon's agar media; 2-5 on Corn meal agar, Czapek's agar and Malt salt agar media; 3-7 on Potato dextrose agar medium and 2-7 on Richard's agar medium. Septation in conidia was also observed variable on different types of media. Transverse septa varied from 1-5 to 2-8 viz; 1-4; 1-5; 2-7 and 2-8 on Corn Meal agar medium; Asthana and Hawker's and Brown's agar media; Coon's agar; Czapek's agar; Leaf decoction agar; Malt salt agar and Sabouraud's agar media and Potato dextrose and Richard's agar media respectively, while longitudinal septa varied from 0-3 to 0-6 viz.; 0-3 on Asthana and Hawker's agar; Brown's agar, Coon's agar and Corn meal agar media; 0-4 on Czapek's agar; Leaf decoction; Malt salt and Sabouraud's agar media; 0-5 on Richard's agar medium and 0-6 on Potato dextrose agar medium. Conidia, were also found varied in size from $7.0-35.0 \times 3.0-13.80 \mu$ in size on different types of media.

The beaks were found septate, and varied in size from $10.0-78.40 \times 1.20-6.40 \mu$. The transverse septa varied from 0-1 on Asthana and Hawker's agar, Czapek's agar, Leaf decoction agar, Malt extract agar and Sabouraud's

agar media and 0-2 on Brown's agar, Coon's agar, Corn meal agar, Potato dextrose agar and Richard's agar media.

The beaks were also recorded as olive green; olive buff to brown; light brown; olive grey; dark olive brown; olive to dark brown and light olive buff on Asthana and Hawker's agar medium; Brown's agar medium; Coon's agar and Richard's agar media; Corn meal agar medium; Czapek's and Sabouraud's agar media; Leaf decoction agar medium; Malt salt agar medium and Potato dextrose agar medium respectively. The beaks were reported as cylindrical on corn meal, Czapek's, Leaf decoction; Richard's and Sabouraud's agar media and conical on Asthana and Hawker's; Brown's Agar; Coon's, Malt salt agar and Potato dextrose agar media.

Chlamydospores were found terminal as well as intercalary varying in size from 4.70- 22.80 μ in diameter on all the different types of media but varying in size as dark brown; dark olive green; dark olive brown; olivaceous to dark brown; olive buff to brown; olive buff; light brown and olive grey on Czapek's and Sabouraud's agar medium, Asthana and Hawker's agar, Leaf decoction agar, Malt salt agar; Brown's agar, Potato dextrose agar; Coon's agar and Richard's agar media and Corn meal agar medium respectively.

The pathogen was also grown on different non- synthetic, semi-synthetic and synthetic media to select the ideal medium form, carrying out for further physiological activities of the pathogen. Out of ten liquid media studied Potato dextrose medium was found to boost the fungal growth weighing 468.30 mg. mycelial growth as well as excellent sporulation followed by Richard's medium. Brown's medium yielded poorest growth and poor sporulation.

In solid states of the media findings showed a trend almost similar to that found with their liquid forms, ofcourse there are some variations in the order of their superiority. In present investigation Potato dextrose agar medium took first place instead of Richard's medium in terms of mycelial growth. Almost close correlation, was observed between growth and

sporulation in dry weight and linear growth of pathogen. Potato dextrose agar medium, was selected as a basal medium for physiological and enzymatic studies due to uniform best growth and sporulation. Some variations in cultural and morphological characters, were recorded on different culture media but significant differences on morphological characters were not observed.

The test organism could grow on a wide range of temperatures of 5°C to 50°C viz; 5°C (T-1), 10°C (T-2), 15°C (T-3), 20°C (T-4), 25°C (T-5), 30°C (T-6), 35°C (T-7), 40°C (T-8), 45°C (T-9) and 50°C (T-10). The optimum range being 30°C (T-6) and 35°C (T-7). The best growth and sporulation was recorded at 30°C (T-6), followed by 35°C (T-7). Sporulation, was also recorded as excellent at 25°C (T-5), 30°C (T-6), and 35°C (T-7), good at 20°C (T-4) and 40°C (T-8), fair at 15°C (T-3) and poor at 5°C (T-1) and 45°C (T-9). The pathogen failed to sporulate at 5°C (T-1) and 50°C (T-10).

The pathogen was able to grow on pH ranged from 2.50 to 12.0 viz; P-1 (2.50), P-2 (3.0), P-3 (3.50), P-4 (4.0), P-5 (4.50), P-6 (5.0), P-7 (5.50), P-8 (6.0), P-9 (6.50), P-10 (7.0), P-11 (7.50), P-12 (8.0), P-13 (8.50), P-14 (9.0), P-15 (9.5), P-16 (10.0), P-17 (10.50), P-18 (11.0), P-19 (11.50) and P-20 (12), but sporulated between the range of pH 3.50 (P-3) to pH 10.50 (P-17).

The maximum growth weighing 495.0 mg was observed at pH 6.50 (P-9), which was referred to as optimum pH. Excellent sporulation, was observed as P-9 (pH 6.50); followed by P-10 (pH 7.0); good at P-11 (pH 7.50) and P-12 (pH 8.0); fair at P-5 (pH 4.50), P-6 (pH 5.0), P-7 (pH 5.50) P-8 (pH 6.0), P-13 (pH 8.50), P-14 (pH 9.0) and P-15 (pH 9.50) and poor at P-3 (pH 3.50), P-4 (pH 4.0), P-16 (pH 10.0) and P-17 (pH 10.50). The pathogen failed to sporulate on P-1 (pH 2.50), P-2 (pH 3.0), P-18 (pH 11.0), P-19 (pH 11.50) and P-20 (pH 12.0). The pH, altered the pH of medium towards neutrality side. It was also observed that reaction of medium tended towards alkaline in cases, where the pH was on acidic side and vice-versa in the cases, where the media, was adjusted at P-14 (pH 9.0) to P-16 (pH 10.0) initially.

The studies on production of enzymes by the fungus *in vitro*, revealed that it produced Cellulase (CX), Polygalacturonase (PG) and Polymethylgalacturonase (PMG) enzymes. It was found that the activity of the enzymes was comparatively more in the medium supplemented with Carboxymethylcellulose (CMC), Sodium polypectate and Citrus pectin respectively; which play an important role in pathogenesis. In *in vivo* studies, it was found that activity of Cellulase (CX), Polygalacturonase (PG) and Polymethylgalacturonase (PMG) enzymes took place in the diseased leaves inoculated with pathogen, *Alternaria alternata* (Fries.), Keissler and no enzymatic activity was produced in healthy leaves.

As regards the susceptible age of the host, it was observed that the pathogen may cause the disease at any stage of plant growth but the maximum susceptibility was observed in the plants, which attained the age of 50 days, followed by 60, 40 and 70 days old plants. The minimum disease intensity, was recorded from 10 days old plants. It was also concluded that plants were susceptible to disease at the age of 40-60 and 50-70 days particularly at 50 days. The susceptibility of plants towards disease decreased with the increasing age of plants and found almost as traces at the age of 90 days and onward. In general the susceptibility of plants to the disease gradually was found decreased below or above 50 days old plants.

The environmental factors like atmospheric temperature, relative humidity and rainfall, were proved to have profound influences on the disease incidence. The disease exhibited firstly its appearance in second week of August in both the years 2001 and 2002 and increased gradually. The maximum disease development 38.84 per cent and 35.67 per cent was recorded in the second week of August during both the years 2001 and 2002 respectively, when the average temperature was 29.05°C and 28.70°C; relative humidity 86.70 per cent and 84.25 per cent and rain fall 0.29 mm. and 11.48 mm. respectively. A trend of decline of disease severity was also recorded with lowering down the temperature and relative humidity during

the month of May and finally by the third week of October, when both the atmospheric temperature and relative humidity, were unfavourable. In general the disease intensity decreased with the increase in temperature, whereas increased with the increase in relative humidity in both the years 2001 and 2002. The effect of rain fall, however was relatively more important in epidemiology of the disease as compared to distribution of rainfall 5-6 days a week accompanied by a cloudy weather.

In order to find out the best carbon source for the growth and sporulation of the pathogen, thirteen different carbon compounds were tested. Sucrose supported the best growth of the pathogen followed by galactose, maltose, raffinose, dextrose, mannose, fructose and xylose, which supported good growth. Fair growth, was observed on sorbitol, lactose and mannitol, whereas poor on dextrin and rhamnose. All the sources of carbon were significantly superior to control, which exhibited minimum growth of pathogen. Investigation on sporulation of the pathogen on different carbon sources, revealed that sucrose, maltose and raffinose supported excellent sporulation. Good sporulation was recorded on galactose dextrose, mannose, fructose, xylose and mannitol; fair on sorbitol, lactose and dextrin, whereas poor sporulation recorded on rhamnose. No sporulation was recorded in case of control.

With a view to find out the best nitrogen source for the growth and sporulation of the pathogen, *A. alternata*, twelve organic and inorganic nitrogenous compounds, were tested in the present study. Of these, peptone supported the best growth of fungus, whereas good growth and obtained on ammonium chloride, ammonium nitrate, calcium nitrate, potassium nitrate and sodium nitrate; fair growth on ammonium acetate, ammonium oxalate and ammonium sulphate and poor on ammonium carbonate, thiourea and urea. Excellent sporulation was recorded on peptone and sodium nitrate; good on ammonium nitrate, calcium nitrate and potassium nitrate; fair on ammonium chloride and ammonium oxalate and poor on ammonium

carbonate, ammonium sulphate and urea, whereas no sporulation was observed in case of the urea and control.

In respect of the effect of nitrogen, phosphorus and potash on disease severity, it was recorded that 60 Kg. P_2O_5 + 40 Kg. K_2O , were more effective in reducing the disease intensity as compared to 60 Kg. N + 60 Kg. P_2O_5 + 40 Kg. K_2O per hectare, whereas when 120 Kg. N per hectare was given alone. No work appears to have been done on the host nutrition in respect of the incidence of leaf spot of Dolichos bean caused by *A. alternata*.

Changes in biochemical constituents viz; Wax, Chlorophyll, Polyphenol, Reducing and Non-reducing sugars, Nitrogen, Phosphorus, Potassium and Sulphur, were also studied in the healthy and diseased leaves of Dolichos bean at different stages i.e. 40 days and 70 days by inoculating with *Alternaria alternata*. Considerable changes were observed in comparison to healthy leaves. It was also observed that contents of wax, chlorophyll a and b, polyphenols, reducing and non-reducing sugars and nitrogen, were comparatively decreased in both the categories of inoculated leaves in descending order by utilizing them for its nutritional requirement or by destroying them through reaction. The remarkable changes regarding reduction in amount of wax, chlorophyll a and b, polyphenols and reducing sugars in the necrotic tissues of the leaves after 70 days of inoculation were observed but no changes were recorded in contents of phosphorus, potash and sulphur.

The investigation on mode of survival of the fungus revealed that the pathogen remained viable in soil, seeds and plant debris from November to June till the next sowing season. In tested seeds soil and diseased plant debris, were observed as virulent to serve as a source of primary inoculum for the pathogen. Secondary spread of disease was observed to be caused by conidia produced on the diseased spots of infected leaves and transmitted through air.

For screening the source of resistance against leaf spot of Dolichos bean

93 germplasms / cultures, were examined under natural and artificial conditions in order to examine their reactions to pathogen. In natural conditions, six germplasms / cultures viz; Arka Vijaya, Cultures 7703, 7008-B and 9101, JDL-37 and Todi-125-136 were found Tolerant being Disease Free (F); Five germplasms/cultures viz; Altapati, Culture-7022, JDL-17, Pusa Early Prolific and Rajani were recorded as Resistant (R) having the disease intensity varied from 1.45 per cent to 2.75 per cent; twelve germplasms/cultures viz; Arka Jai, Cultures-6802, 7001, 7103, 7301, 8001, 8002, 8403, HA-3, HD-4, HD-81 and JDL-85, were recorded as Moderately Resistant (MR) having the disease intensity varied from 6.25 per cent to 9.13 per cent; thirty germplasms/cultures viz; Culture-5508, 6001, 6201, 6317, 6804, 7008-A, 7010, 7024, 7027, 7210, 7601, 7701, 7705, 7708, 8003, 8004, 8005, 8405, 9102, 9108, 9109, 9110, 9113, 9117, 9118, DPL-1, Goya, HD-10, HD-66, HD-104 and Todi-21, were found Moderately Susceptible (MS) having the disease intensity varied from 11.72 per cent to 18.29 per cent; twenty six germplasms/ cultures viz; Cultures-6022, 6023, 6701, 6801, 7006, 7007, 7012, 7015, 7016, 7019, 7020, 7101, 7205, 7206, 7603, 7702, 7710, 7711, 8101, 8401, 9104, 9105, 9114, 9116, DB-1 and Hatikan, were found Susceptible (S), having the disease intensity varied from 20.54 per cent to 29.27 per cent and thirteen germplasms/cultures viz; Cultures-6009, 6014, 6019, 7005, 7015-B, 7020-A, 7023, 7501, 7604, HD-93, JDL-79, Kalyanpur Type-1 and Kamgranj Selection-2, were found Highly Susceptible (HS), having the disease intensity varied from 51.49-62.38 per cent.

Further in artificial epiphytotic conditions of seeds and plant inoculation for their reaction to pathogen during Kharif season in the year 2001, the seeds of 23 germplasms/cultures, which were found Disease Free (F), Resistant (R) and Moderately Resistant (MR), were further examined. In seed inoculation test none of the Dolichos bean germplasms/cultures, was found to be immune and resistant. Out of 23 germplasms/cultures tested in seed inoculation test, ten germplasms/cultures viz., Arka Vijai, Cultures 6802, 7022, 7301, 8002 and 9101, HA-3, HD-81, Rajani and Todi-125-136 were

found Moderately Resistant (MR), having the disease intensity varied from 5.80 to 9.45 per cent; seven germplasms/cultures viz., Altapati, Arka Vijaya cultures 6802, 7708-B, DDL-37, HD-4 and JDL-17 were found Moderately Susceptible (MS) having the disease intensity varied from 10.36 to 18.59 per cent and remaining six germplasms/cultures (Cultures 7103, 8001, 8403, 7703, JDL-85 and DL-37 were found Susceptible (S) having the disease intensity varied from 23.82 per cent to 28.80 per cent, whereas in inoculation of potted plants, nine germplasms/cultures Arka Jai, Culture-7001, 7022, 9101, DDL-37, HD-4, JDL-85, Rajani and Todi-135-136 were found Moderately Resistant (MR), having the disease intensity varied from 6.28 per cent to 9.16 per cent; nine germplasms/cultures Altapati, Arka Vijaya, Cultures, 6802, 7301, 8001, 8002, HA-3, JDL-17 and JDL-37 were found Moderately Susceptible (MS), having the disease intensity varied from 11.92 per cent to 18.29 per cent and remaining five germplasms/cultures Cultures 7103, 7793, 7708-B, 8403 and HD-81, were found Susceptible (S) having the disease intensity varied from 21.54 per cent to 29.67 per cent.

Host range study revealed that the pathogen was able to infect the wide range of 70 host plants, both cultivated and wild belonging to 19 different families viz; Apocynaceae, Aracaceae, Chenopodiaceae, Asteraceae, Brassicaceae, Cucurbitaceae, Euphorbiaceae, Poaceae, Fabaceae, Labiatae (Lamiaceae), Liliaceae, Linaceae, Malvaceae, Myrtaceae, Papaveraceae, Pedaliaceae, Rosaceae, Solanaceae, and Apiaceae (Umbelliferae). Out of these 70 plants *Abelmoschus esculentus*, *Abutilon indicum*, *Althaea rosea*, *Allium cepa*, *Argemone mexicana*, *Avena sativa*, *Arachis hypogea*, *Brassica campestris*, *B. Campestris* var. *dichotoma*, *B. juncea*, *B. oleracea* var. *botrytis*, *B. oleracea* var. *capitata*, *B. oleracea* var. *gongylodes*, *Carissa carandus*, *Carthamus tinctorius*, *Cajanus cajan*, *Capsicum annuum*, *Chenopodium album*, *Chrysanthemum indicum*, *Colocasia antiquorum*, *Coriandrum sativum*, *Crotalaria juncea*, *Cucurbita maxima*, *Cynodon dactylon*, *Dahlia* sp, *Datura alba*, *Gossypium* sp., *Glycine max*, *Hibiscus rosa-sinensis*, *Hordeum vulgare*, *Linum usitatissimum*, *Lagenaria vulgaris*, *Lycopersicum esculentum*,

Luffa cylindrica, *Ocimum sanctum*, *Pisum sativum*, *Pennisetum typhoides*, *Raphanus sativus*, *Ricinus communis*, *Saccharum officinarum*, *Sesamum indicum*, *Sida acuta*, *Sorghum vulgare*, *Solanum nigrum*, *S. melongena*, *S. xanthocarpum*, *S. tuberosum*. *Tagetes erecta* and *Triticum aestivum*, belonging to fifteen different families were found infected under artificial conditions of inoculation with spore-cum-mycelial suspension of the pathogen. It was found that the pathogen could infect the monocotyledonous and dicotyledonous plants having wide host range.

To select the suitable fungitoxicant for the control of the disease under field conditions the efficacy of twenty six fungitoxicants, Benzene (Halogenated organic compound), Inorganic Copper compound, Dithiocarbamate, Heterocyclic nitrogenous compound, Organomercury, Quinone, Systemic inorganic Sulphur compound and an Antibiotic viz; Agrosan G.N.; Bavistin, Benlate, Blitox-50, Brassicol, Calixin, Captafol, Captan, Cersan (Dry), Dichlone, Dithane M-45, Dithane Z-78, Duter, Emisan-6, Ferbam, Foltaf. 80-W, Karathane, Kavach, Hexaferb, Pancotine, Ridomil, Spergon, Suflex, Thiram, Vitavax and Ziram as well as an antibiotic (Aureofungin), were tested in the laboratory on the effect of growth of fungus, *Alternaria alternata* revealed that twelve of them namely Agrosan G.N.; Aureofungin, Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf 80-W, Karathane, Kavach, Pancotine, Thiram and Vitavax proved to most effective as inhibited the growth completely. The rest fungitoxicants, were found partially effective. Dithane M-45 caused the highest inhibition of fungal growth followed by Dichlone. The other fungicides in descending of efficacy, were suflex, Calixin, Blitox-50, Brassicol, Ridomil, Benlate, Ferbam, Ziram, Spergon, Bavistin, Hexaferb and Dithane Z-78. Dry weight of fungal mycelium in different concentrations of 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent and 0.60 per cent, were significantly better in performance in comparison to control. A dosage response in different concentrations viz; 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent and 0.60 per cent of fungitoxicants with reference to the

pathogenic effect viz., light yellow lesion (LY), Yellowish brown lesion (YB), Light yellow brown lesion (LB), Extensive brown lesion (EB) and Phytotoxic effect (PE) on the plants were also studied. Out of the 25 fungitoxicants and an antibiotic (Aureofungins) tested Agrosan G. N., Ceresan (Dry) and Emisan-6, belonging to Organomercury, Duter, Karathane and Thiram belonging to Dithiocarbamate, Aureofungin (Antibiotic), Captan and Foltaf 80-W (Captafol) belonging to Hetrocyclic nitrogenous compound and Panacotine and Vitavax belonging to Systemic group were most effective as they inhibited the growth of fungus and did not produce any pathogenic effects. Other fungicides viz.; Calixin (Systemic) and Suflex (Inorganic sulphur), were also found effective in checking the growth of fungus and superior to the remaining fungicides tested. These fungitoxicants were found to be most effective as they inhibited the growth of fungus, when tested the different concentrations of 0.05 per cent, 0.10 per cent, 0.15 per cent, 0.20 per cent, 0.40 per cent, and 0.60 per cent, in respect of dry weight of mycelium. The other fungicides viz; Calixin (Systemic) and Suflex (Inorganic sulphur), were found also effective in arresting the growth of fungus upto 85.70 per cent at 0.05 per cent, 88.60 per cent at 0.10 per cent, 88.98 per cent at 0.15 per cent, 70.48 per cent at 0.20 per cent, 91.20 per cent at 0.40 per cent and 95.69 at 0.60 per cent and 88.91 per cent at 0.05 per cent, 92.44 per cent at 0.10 per cent. 93.41 per cent in 0.15 per cent. 91.33 per cent in 0.20 per cent, 94.85 per cent in 0.40 per cent and 95.79 per cent in 0.60 per cent respectively. The significant variations in inhibition in hyphal dry weight with respect to treatment of different fungitoxicants as well as different concentrations, were recorded. Apart from this these fungitoxicants were also found effective as did not produce any pathogenic effects but Brassicol (Benzene), Calixin (Systemic) and Suflex (Inorganic sulphur), were also found effective in producing Light yellow lesions (LY) only in different concentrations of fungitoxicants and established better in comparison to the remaining fungitoxicants tested except Brassicol (Benzene) in 0.30 per cent, where no pathogenic effect were shown. The number of germ tubes varied

from 5-38, 6-31 and 2-29 in 0.002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants, Minimum 6, 5, and 2 germ tubes were produced in treatment with Dithane M-45 (Systemic) in 0.0002, 0.0004 and 0.0006 per cent concentrations respectively, while maximum 37, 30 and 29 germs tubes in the treatment of Calixin (Systemic) and in treatment with other fungicides 10, 28, 36, 32, 11, 12, 28, 16, 30, 20, 27 and 20, 18, 23, 27, 28, 9, 11, 20, 14, 25, 18, 28 and 17, 6, 28, 27, 27, 21, 3, 16, 11, 23, 18, 28 and 14 germ tubes were found in 0.0002, 0.0004 and 0.0006 per cent concentrations. They were also effective as fungal spores failed to germinate, when fungitoxicants, were applied in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations but Dithane M-45 (Dithiocarbamate) and Vitavax were less effective as produced minimum spore germination and proved better in comparison to other fungitoxicants. As for as the length of germ tube is concerned, varied from 0.005 to 0.018; 0.007 to 0.016 per cent and 0.006 to 0.014 per cent in 0.002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of different fungitoxicants respectively. The minimum germ tube length measuring 0.005 mm., 0.007 mm. and 0.006 mm., were recorded in treatment with Bavistin (Systemic) in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants respectively, whereas maximum number of germ tubes measuring length 0.017 mm., 0.014 mm. and 0.013 mm. with the treatment of Calixin (Systemic) in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations respectively. The inhibition percentage over control varied from 24.82 per cent to 68.72 per cent, 34.74 per cent to 74.28 per cent and 45.48 per cent to 78.19 per cent in 0.0002 per cent, 0.0004 per cent and 0.0006 per cent concentrations of fungitoxicants respectively. The inhibition in all the three parameters in spore germination, germ tube length and number of germ tubes, produced by spores were pronounced in higher concentrations of fungitoxicants with significant differences. Dithane M-45 (Dithiocarbamate) caused appreciable degree of inhibition in respect of germination of spore, germ tube number and germ tube length. The germ tube length was inhibited up to 78.19 per

cent at higher concentrations. At lower concentration, Blitox-50 (Inorganic Copper) and Calixin (Systemic), were not proved effective but effective at higher concentration in inhibiting spore germination.

The efficacy of selected seed dressing fungitoxicants viz., Agrosan G. N., Aureofungin, (Antibiotic), Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80 W (Captafol) Karathane, Kavach, Pancotine, Thiram and Vitavax as seed dressers proved effective in laboratory, were evaluated on seed treatments for studying their effects on seed germination and seed borne infection *in vitro* and it was found that all of them improved germination. The highest germination was recorded in seeds treated with Captan (Heterocyclic nitrogenous compound) Ceresan (Dry organomercury), Thiram (Dithiocarbamate), Agrosan G. N. (Organomercury), Aureofungin (Antibiotic), Emisan-6 (Organo-mercury), Duter (Dithiocarbamate), Pancotine (Systemic), Karathane (Dithiocarbamate), Foltaf 80-W (Heterocyclic nitrogenous compound), Kavach and Vitavax (Systemic). The seed treatment with Ceresan (Dry) resulted in elimination of the fungus but the treatment with Thiram (Dithiocarbamate), Captan (Heterocyclic nitrogenous compound) and Agrosan G.N. (Organomercury), expressed their broad spectrum in nature in order mentioned and these were par in efficacy. It was also recorded that lowest infection 1.29 per cent was found in treatment with Captan. The maximum 14.55 per cent infection was recorded in treatment with Vitavax followed by Pancotine (Systemic), Thiram (Dithiocarbamate), Emisan-6 (Organomercury), Duter (Dithiocarbamate), Aureofungin (Antibiotic), Agrosan G.N. (Organomercury), Foltaf 80W (Heterocyclic nitrogenous compound), Karathane (Dithiocarbamate), Kavach and Captan (Heterocyclic Nitrogenous compound). Ceresan (Dry, organomercury), was found to be best one as it completely eliminated the infection. In general all the fungicides examined, were found significant over control in reducing seed borne infection.

The same seed dressing fungitoxicants were tested in pot experiments also ascertaining their effects on seed germination and seedling infection. The highest seed germination 95.47 per cent was recorded in treatment with Ceresan (Dry, Organomercury), followed by 92.08 per cent with Captan

(Heterocyclic nitrogenous compound), 91.57 per cent in Thiram (Dithiocarbamate compound) and 90.45 per cent in Agrosan G. N. (Organomercury) 84.78 per cent in Emisan-6 (Organomercury), 83.75 per cent in Duter (Dithiocarbamate), 83.98 per cent in Pancotine (Systemic) 83.79 per cent in Aureofungin (Antibiotic), 81.67 per cent in Karathane (Dithiocarbamate), 79.15 per cent in Kavach and 78.60 per cent in Vitavax, while comparatively poor germination was noticed in treatment with Pancotine (Systemic), Karathane (Dithiocarbamate), Vitavax (Systemic) and Foltaf-80 W (Heterocyclic nitrogenous compound). The complete control of seedling infection in treatment with Ceresan (Dry, Organomercury), Thiram (Dithiocarbamate) and Vitavax (Systemic) had no infection. Captan (Heterocyclic nitrogenous compound) and Agrosan G. N., were found superior in elimination of seedling infection. Thus seed dressing fungitoxics belonging to Heterocyclic nitrogenous compound (Captan), Organomercurials (Agrosan G. N. and Ceresan) compound groups were most efficacious than fungitoxics belonging to Dithiocarbamate, Copper, Sulphur and Quinone compounds of Systemic nature controlling seeds borne infection.

In studies regarding the control of disease in the field, twelve different fungitoxics viz., Agrosan G. N., Captan, Ceresan (Dry), Duter, Emisan-6, Foltaf-80 W, Karathane, Kavach, Pancotine, Thiram and Vitavax as well as an antibiotic (Aureofungin), which were found completely or partially effective in bioassay test, were evaluated under pot and field in the years 2001 and 2002 in order to select out a suitable fungitoxicant reducing the incidence of disease and boosting seed yield. The best control of disease was obtained by application of Captan (Heterocyclic nitrogenous compound) followed by Thiram (Dithiocarbamate), Ceresan (Dry), Agrosan GN (organomercury), Karathane (Dithiocarbamate), Kavach, Duter (Dithiocarbamate), Aureofungin (Antibiotic), Pancotine (Systemic), Emisan-6 (Organomercury), Vitavax (Systemic), and Foltaf 80-W (Heterocyclic nitrogenous compound), differed significantly in the year 2001 and 2002 in pot experiment while in Field experiment followed by Thiram

(Dithiocarbamate), Vitavax (Systemic), Ceresan (Dry organomercury), Agrosan G.N. (Organomercury), Aureofungin (Antibiotic), Duter (Dithiocarbamate), Emisan-6 (Organomercury), Pancotine (Systemic), Foltaf 80-W (Heterocyclic nitrogenous compound), Karathane (Dithiocarbamate) and Kavach differed significantly in the year 2001 and 2002 and were significantly superior over control. Foltaf 80-W and Kavach, fungitoxicants were found poorest amongst all the fungicides controlling the disease in both the years, 2001 and 2002 in pot and field experiments respectively. Further it was also recorded the plots treated with Captan exhibited significantly better yield 31.34 and 29.08 gms. per plant than control gms. weighing 19.62 gms. and 19.82 gms. and 19.25 gms. and 17.32 gms. than control weighing 15.87 and 14.75 gms. per plate, in both the years 2001 and 2002 in pot and field experiments respectively. The next in effectiveness was Thiram followed by Agrosan G.N.; Ceresan (Dry), Aureofungin (Antibiotic), Emisan-6, Pancotine, Duter, Karathane and Kavach in Pot experiments and Thiram, Ceresan (Dry), Agrosan, G.N., Pancotine, Duter, Emisan-6, Vitavax, Karathane, Kavach, Aureofungin (Antibiotic) and Foltaf 80-W in field experiments respectively in the years 2001 and 2002. Lowest yield weighting 19.94 gms. and 20.35 and 16.08 gms. in pot experiment and 13.42 gms. per plant in field experiments in treatment with Vitavax and Foltaf 80-W in both the years. Complete control of the disease was not achieved by any of the fungitoxicants tested but significantly in the yield establishing a good correlation between Leaf infection and yield, which were lowered according to the severity.

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Chapter - VII

APPENDIX

Table - XXXXI
List of Abbreviations

Abbreviations	Full Form
°C	Centigrade
cm.	Centimeter
g.	gram
mm.	milimeter
LB	Light Brown Lesions
LY	Light yellow Lesions
YB	Yellowish Brown Lesions
EB	Extensive Brown Lesions
PE	Phytotoxic Effect
P ₁	2.50 pH
P ₂	3.00 pH
P ₃	3.50 pH
P ₄	4.00 pH
P ₅	4.50 pH
P ₆	5.00 pH
P ₇	5.50 pH
P ₈	6.00 pH
P ₉	6.50 pH
P ₁₀	7.00 pH
P ₁₁	7.50 pH
P ₁₂	8.00 pH
P ₁₃	8.50 pH
P ₁₄	9.00 pH
P ₁₅	9.50 pH
P ₁₆	10.00 pH
P ₁₇	10.50 pH
P ₁₈	11.00 pH
P ₁₉	11.50 pH
P ₂₀	12.00 pH
T ₁	5°C
T ₂	10°C
T ₃	15°C
T ₄	20°C

T ₅	25°C
T ₆	30°C
T ₇	35°C
T ₈	40°C
T ₉	45°C
T ₁₀	50°C
μ	micron
—	Absent/very poor
+	Poor
++	Fair
+++	Good
++++	Excellent

Details of Infection

—	No infection
+	Up to 50% leaf area affected
++	5-10% Leaf area affected.
+++	11-20% Leaf area affected.
++++	20-30% Leaf are affected.
+++++	Above 30% leaf area affected.
0	No infection.
1	Upto 5.0 per cent leaf area affected.
2	Upto 10.0 per cent leaf area affected.
3	Upto 20.0 per cent leaf area affected.
4	Upto 30 per cent leaf area affected.
5	Upto 40.0 per cent leaf area affected.



Chapter - VIII
BIBLIOGRAPHY

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